4-RESULTS

4.1- Biological studies:

To study the effect of the present IGRs on some biological aspects of treated stages and sub-sequent developmental stages of *Spodoptera littoralis*, the experiments were carried out on the 2nd and 4th larval instars. Castor bean leaves, *Ricinus communus*, were dipped in different concentrations of the tested IGRs on which starved larvae feed for 24 hour

4.1.1. Effect of Cascade on some biological aspects of *S. littoralis* treated as 2nd larval instar:

The data obtained in **Table (1)** and **Fig. (1)** showed that the corrected percentages of larval mortality had a positive relationship with the different concentrations of Cascade. The percentages of larval mortality were 30, 60, 80, 95 and 100 % at the Conc.of 0.1, 0.5, 1.0, 1.5 and 2.0 ppm, respectively. The response was dose dependent (i.e. the higher concentration affected more larvae). On the other hand the data obtained in the same **Table** and **Fig**. indicated that there was an inverse relationship between the different concentrations of Cascade and pupation percentages, 70, 40, 20, 5 and 0.0 % at Conc.of 0.1, 0.5, 1.0, 1.5 and 2.0 ppm, respectively, as compared with 100% in the control. While the percentages of pupal mortality was increased with the increase in concentrations. Also the percentages of the adult emergence were decreased with the increasing in concentrations as compared with control.

Total inhibitions of adult emergence were 36.25, 70, 91.25, 96.25 and 100 % at the Conc.of 0.1, 0.5, 1.0, 1.5 and 2.0 ppm, respectively, as compared with 0.0% in the control. The response was dose dependent (i.e the higher concentration induce more inhibition).

The larval and pupal durations were increased with the increasing of concentrations as compared with control, (i.e. the higher concentration induce more prolongation in both larval and pupal durations), as indicated in **Table (1)** and **Fig. (2).**

The fecundity and fertility were decreased as a result of treatment with Cascade. This decrease was negatively correlated with the concentration. On the other hand, the oviposition deterrent index (O.D.I) and percentages of sterility were positively correlated with the concentrations as indicated in **Table (2)** and **Fig. (3)** for instance, (O.D.I) was 1.02, 2.53, 10.19, 12.22 and 0.0 % at the concentrations of 0.1, 0.5, 1.0, 1.5 and 2.0 ppm, respectively. Also, the percentage of sterility was 5.22, 12.8, 28, 36 and 0.0 % at the previous concentrations.

Table (1): The effect of Cascade on biological aspects of cotton leafworm by feeding newly 2nd instar larvae on treated Castor Leave for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0.0	10 ±0.41	100 ±0.0	----- ±0.0	7 ±0.41	100 ±0.0	----- ±0.0
0.1	*** 30 ±0.41	11 ±0.41	*** 70 ±0.41	* 6.25 ±0.25	*** 10 ±0.41	*** 63.75 ±0.25	*** 36.25 ±1.25
0.5	*** 60 ±0.71	11.75 ±0.25	*** 40 ±0.71	*** 10 ±0.41	*** 12 ±0.41	*** 30 ±0.41	*** 70 ±2.04
1.0	*** 80 ±0.41	12.5 ±0.29	*** 20 ±0.41	*** 11.25 ±0.25	*** 12 ±0.41	*** 8.75 ±0.25	*** 91.25 ±1.25
1.5	*** 95 ±0.0	* 13 ±0.91	*** 5 ±0.0	1.25 ±0.25	*** 14 ±0.56	*** 3.75 ±0.25	*** 96.25 ±1.25
2.0	*** 100 ±0.0	8 ±0.71	*** ----- ±0.0	----- ±0.0	*** ----- ±0.0	*** ----- ±0.0	*** 100 ±0.0

Table (2):Effect of Cascade on fecundity, fertility and sterility against adults of cotton leafworm emerged from 2nd larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	$^{+}$O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1250 ±17.68	------ ±0.0	100 ±0.0	------ ±0.0
0.1	1241 ±7.97	1.02 ±0.21	95.5 ±2.05	5.22 ±2.99
0.5	1188 ±8.16	2.53 ±1.01	91.6 ±1	* 12.8 ±0.9
1.0	*** 1019 ±22.63	** 10.19 ±1.47	** 88.35 ±3.04	*** 28 ±3.45
1.5	*** 980 ±45.28	*** 12.22 ±3.01	*** 81.75 ±3.9	*** 36 ±3.8
2.0	*** ------ ±0.0	------ ±0.0	*** ------ ±0.0	------ ±0.0

***Significant at P = 0.05 ** High significant at P = 0.01**
***** Very high significant at P = 0.001 + Oviposition deterrenet index.**

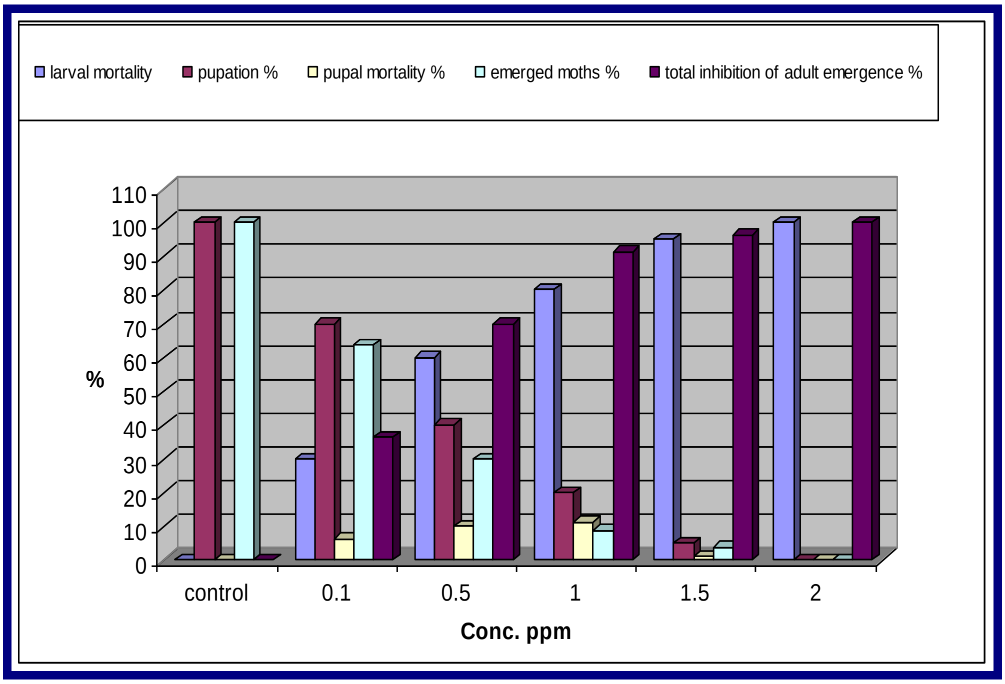


Fig. (1): Effect of Cascade on some biological aspects of *S. littoralis* treated as 2nd larval instar.

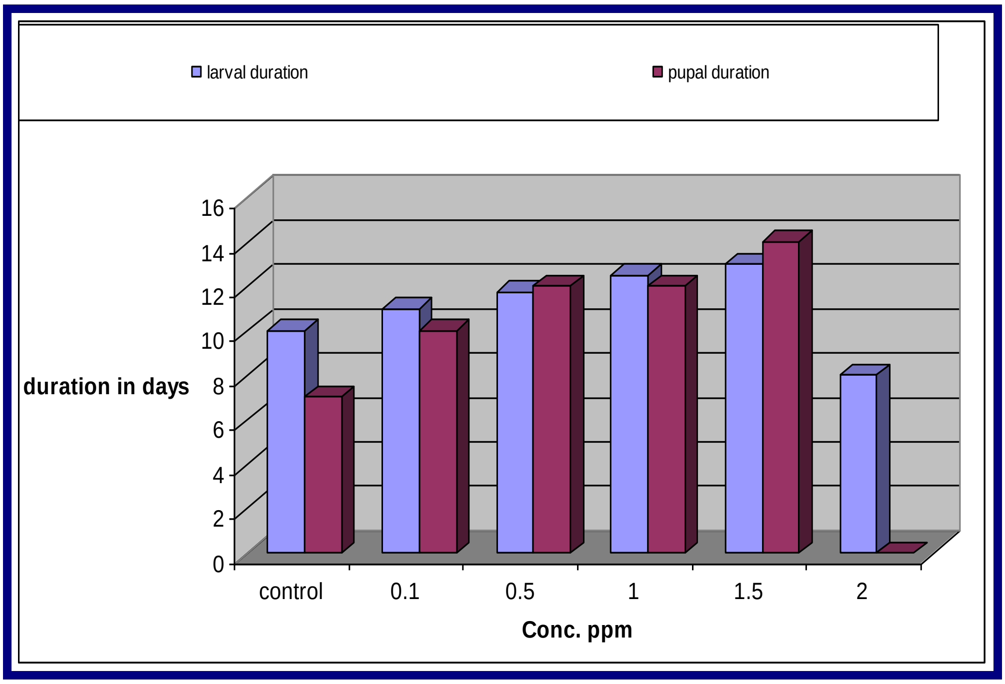


Fig. (2): Effect of Cascade on larval and pupal duration of *S. littoralis* treated as 2nd larval instar.

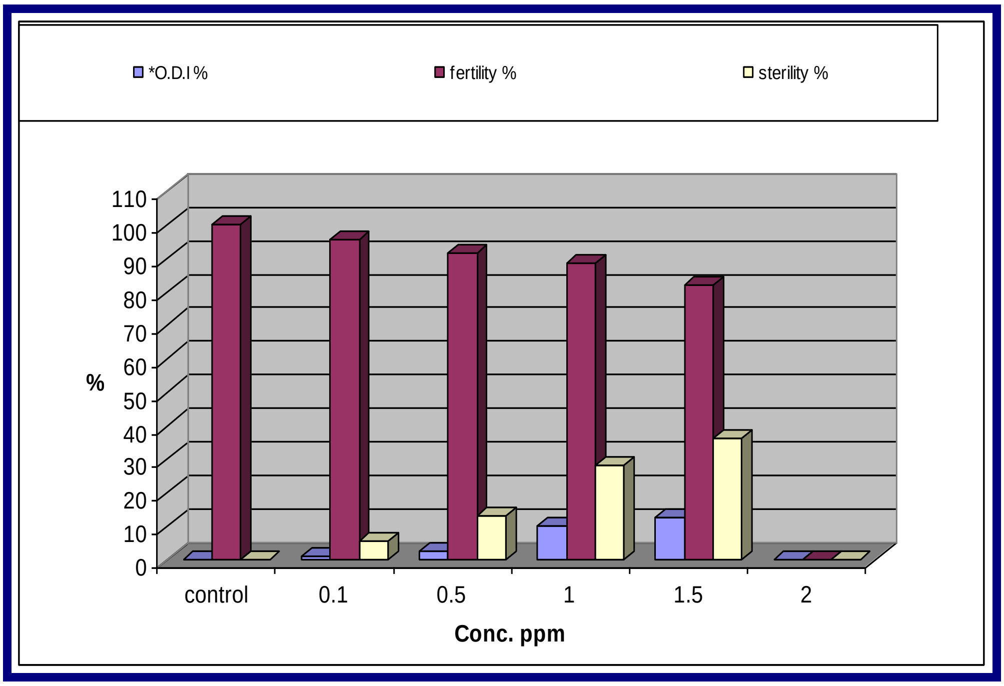


Fig. (3): Effect of Cascade on (O.D.I , fertility and sterility) of *S. littoralis* treated as 2nd larval instar.

4.1.2. Effect of Cascade on some biological aspects of *S. littoralis* treated as 4th larval instar:

Table (3) and **Fig. (4)** declared that there was a highly significant effect on the larval mortality that given in corrected percentages by 10, 32.5, 55, 80 and 100 % at the concentrations of 1, 3, 5, 7 and 9 ppm, respectively. Pupation percentage was greatly reduced 90, 67.5, 45, 20 and 0.0 % as compared with 100 % in control, while the percentages of pupal mortality were increased with the increase in concentrations. The reduction in the adult emergence percentages was increased with the increasing in the concentrations, total inhibitions of adult emergence were 12.5, 45, 71.25, 90 and 100 % at the conc.of 1, 3, 5, 7 and 9 ppm, respectively, as compared with 0.0% in the control. The response was dose dependent. It is remarkable that the larval and pupal durations were increased with the increasing of concentrations as compared with control, (i.e. igher concentration induce more prolongation in both larval and pupal durations), as indicated in **Table (3)** and **Fig. (5).**

Data in **Table (4)** and **Fig. (6)** showed the effect of cascade on the fecundity, (O.D.I), fertility and sterility. The fecundity and fertility were decreased. This decrease was negatively correlated with the concentration. On the other hand, the oviposition deterrent index (O.D.I) and percentages of sterility were positively correlated with the concentrations for instance; (O.D.I) was 1.6, 5.1, 7.7 and 11.2 % at the concentrations of 1, 3, 5 and 7 ppm, respectively. Also, the percentage of sterility was 9.08, 18.6, 32.87 and 56.03 % at the previous concentrations.

Table (3): The effect of Cascade on some biological aspects of the cotton leafworm by feeding newly 4th instar larvae on treated Castor Leaves for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0.0	6 ±0.41	100 ±0.0	----- ±0.0	8 ±0.58	100 ±0.0	----- ±0.0
1	** 10 ±0.41	6.5 ±0.29	** 90 ±0.41	2.5 ±0.29	8 ±0.41	*** 87.5 ±0.29	*** 12.5 ±1.44
3	*** 32.5 ±0.29	*8 ±0.41	*** 67.5 ±0.29	** 12.5 ±0.29	10 ±0.41	*** 55 ±0.0	*** 45 ±0.0
5	*** 55 ±0.41	***10 ±0.41	*** 45 ±0.41	***16.25 ±0.62	*11 ±0.41	*** 28.75 ±0.25	*** 71.25 ±1.25
7	*** 80 ±0.41	***12 ±0.41	*** 20 ±0.41	*10 ±0.41	***12 ±0.41	*** 10 ±0.0	*** 90 ±0.0
9	*** 100 ±0.0	*4 ±0.41	*** ----- ±0.0	----- ±0.0	*** ----- ±0.0	*** ----- ±0.0	*** 100 ±0.0

Table (4): Effect of Cascade on fecundity, fertility and sterility against adults of cotton leafworm emerged from 4th larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	+O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1430 ±24.83	------ ±0.0	100 ±0.0	------ ±0.0
1	1385 ±30.48	1.6 ±0.8	93.89 ±2.92	* 9.08 ±2.82
3	** 1290 ±20.41	*** 5.1 ±0.4	* 90.19 ±1.15	*** 18.6 ±1.29
5	***1225 ±30.14	*** 7.7 ±0.6	*** 78.41 ±1.12	*** 32.87 ±0.54
7	*** 1135 ±14.29	*** 11.2 ±0.8	*** 55.45 ±3.35	*** 56.03 ±2.33
9	*** ------- ±0.0	------- ±0.0	*** ------- ±0.0	------- ±0.0

Significant at P = 0.05** * High significant at P = 0.01**

***** Very high significant at P = 0.001** **+ Oviposition deterrenet index.**

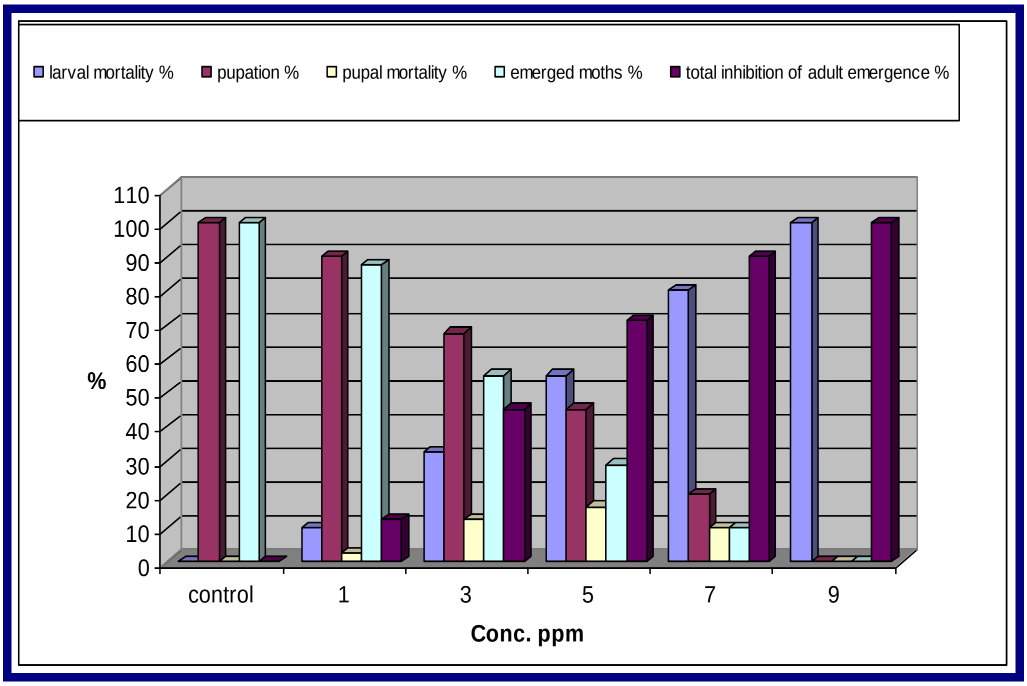


Fig. (4): Effect of Cascade on some biological aspects of

***S. littoralis* treated as 4th larval instar.**

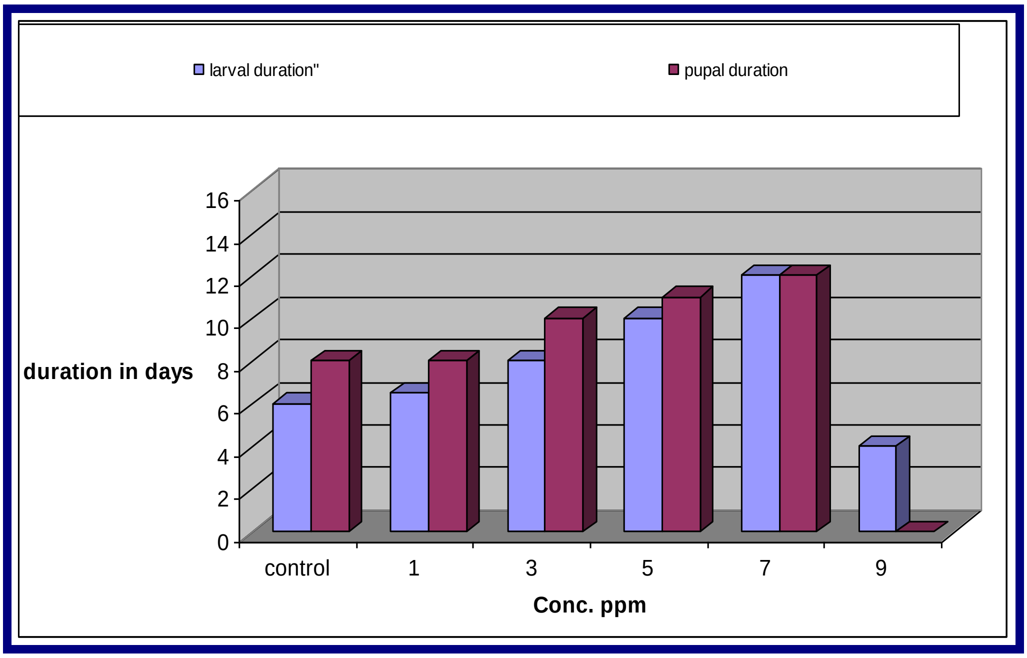


Fig. (5): Effect of Cascade on larval and pupal duration

of *S. littoralis* treated as 4th larval instar.

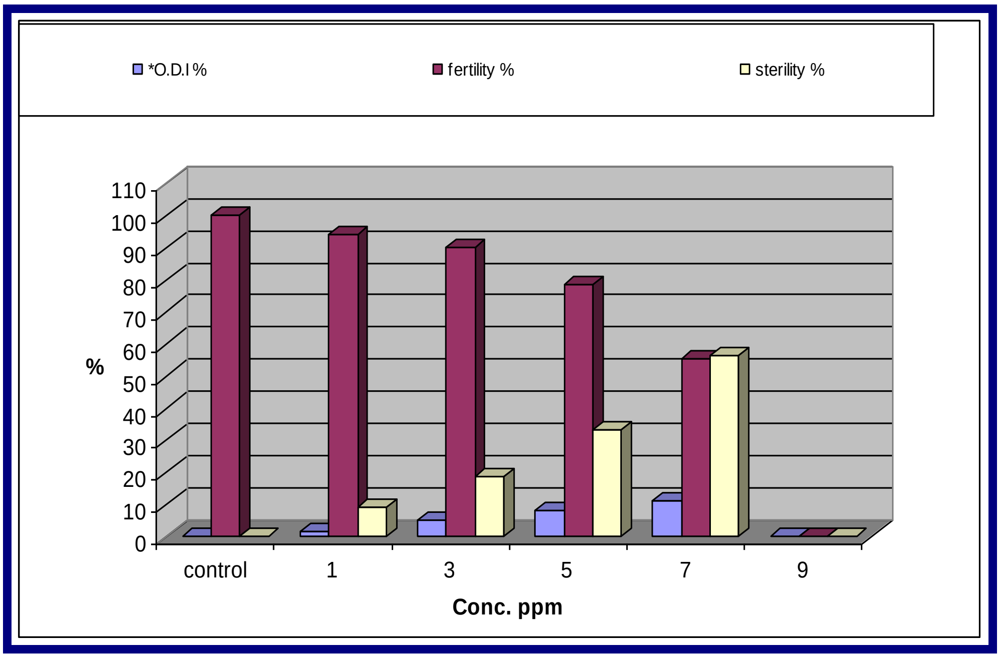


Fig. (6): Effect of Cascade on (O.D.I , fertility and sterility) of *S. littoralis* treated as 4th larval instar.

4.1.3. Effect of Match on some biological aspects of *S. littoralis* treated as 2nd larval instar:

Results recorded in **Table (5)** and **Fig. (7)** showed that there was a significant effect on the larval mortalities that given in corrected percentages by 10, 40, 50, 80 and 100 % at the concentrations of 0.1, 0.5, 1.0, 1.5 and 2.0 ppm, respectively. Also, the pupation percentages were greatly reduced 90, 60, 50 and 20 % as compared with 100 % in control, on the other hand percentages of pupal mortality was increased significantly with the increase in concentrations.

The percentage of the adult emergence was decreased with an increase in the concentrations as indicated by 85, 52.5, 40 and 8.75 % ,respectively, at ascending successive concentrations. The present results indicated that there was a significant inhibition of adult emergence percentages as 15, 47.5, 60, 91.25 and 100 % as compared with control 0.0 %. Larval and pupal durations were increased as shown in the same **Table** and **Fig. (8)**.

On the other hand the results tabulated in **Table (6)** and **Fig. (9)** showed that Cascade induced an reduction in both fecundity and fertility, on the contrast (O.D.I) and sterility showed an positive relationship with the increasing of concentrations.

Table (5): The effect of Match on some biological aspects of the cotton leafworm by feeding newly 2nd instar larvae on treated Castor Leaves for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0.0	10 ±0.41	100 ±0.0	----- ±0.0	7 ±0.41	100 ±0.0	----- ±0.0
0.1	** 10 ±0.0	10 ±0.41	** 90 ±0.0	5 ±0.0	8.5 ±0.29	*** 85 ±0.0	*** 15 ±0.0
0.5	*** 40 ±0.41	11 ±0.58	*** 60 ±0.41	** 7.5 ±0.29	*** 10 ±0.41	*** 52.5 ±0.5	*** 47.5 ±2.5
1.0	*** 50 ±0.58	12 ±0.58	*** 50 ±0.58	*** 10 ±0.41	*** 11 ±0.58	*** 40 ±0.41	*** 60 ±2.04
1.5	*** 80 ±0.41	* 13 ±0.41	*** 20 ±0.41	*** 11.25 ±0.25	*** 13 ±0.41	*** 8.75 ±0.25	*** 91.25 ±1.25
2.0	*** 100 ±0.0	8 ±0.41	*** ----- ±0.0	----- ±0.0	*** ----- ±0.0	*** ----- ±0.0	*** 100 ±0.0

Table (6): Effect of Match on fecundity, fertility and sterility against adults of cotton leafworm emerged from 2nd larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	+O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1385 ±30.48	------ ±0.0	100 ±0.0	------ ±0.0
0.1	1385 ±30.48	2.55 ±0.73	98.9 ±0.42	4.5 ±1.42
0.5	1275 ±30.62	4.13 ±1.51	*** 94.02 ±1.06	*** 13.4 ±2.28
1.0	*** 1156 ±32.19	*** 9.01 ±2.4	*** 90.08 ±0.43	*** 24.6 ±3.39
1.5	*** 945 ±20.61	*** 18.88 ±0.49	*** 85.18 ±1.49	*** 41.7 ±1.17
2.0	*** ------ ±0.0	------ ±0.0	*** ------ ±0.0	------ ±0.0

Significant at P = 0.05** * High significant at P = 0.01**

***** Very high significant at P = 0.001** **+ Oviposition deterrenet index.**

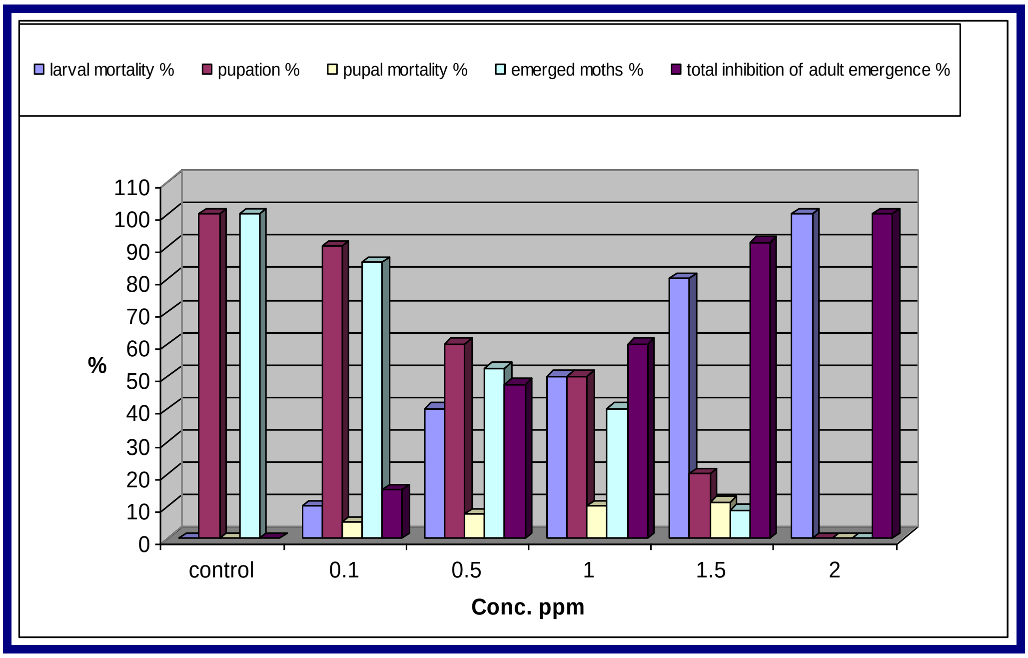


Fig. (7): Effect of Match on some biological aspects of *S. littoralis* treated as 2nd larval instar.

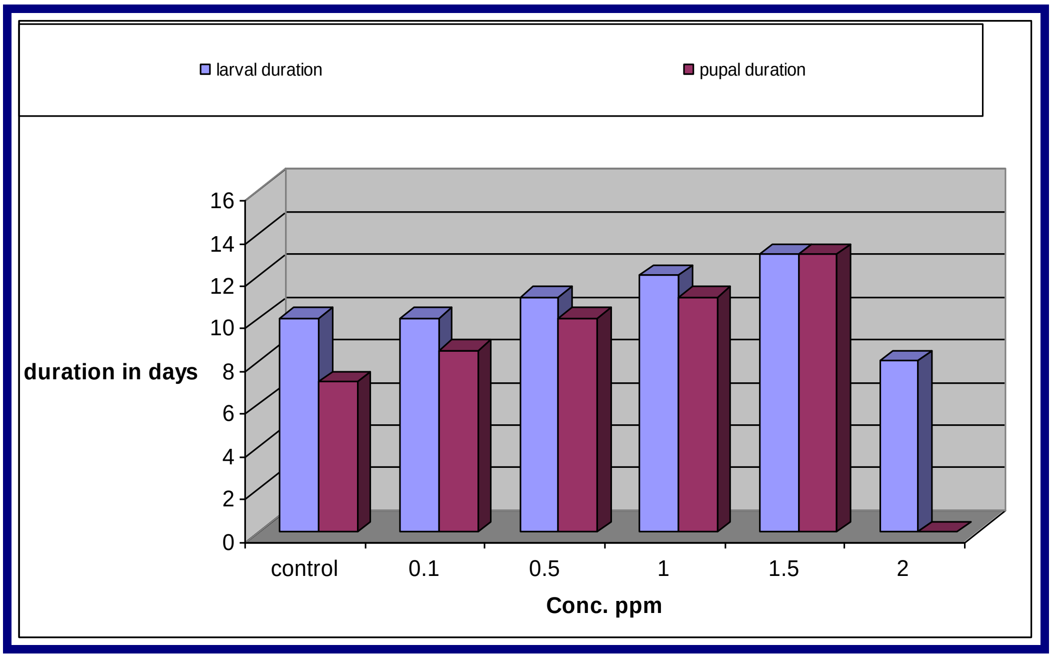


Fig. (8): Effect of Match on larval and pupal duration of *S. littoralis* treated as 2nd larval instar.

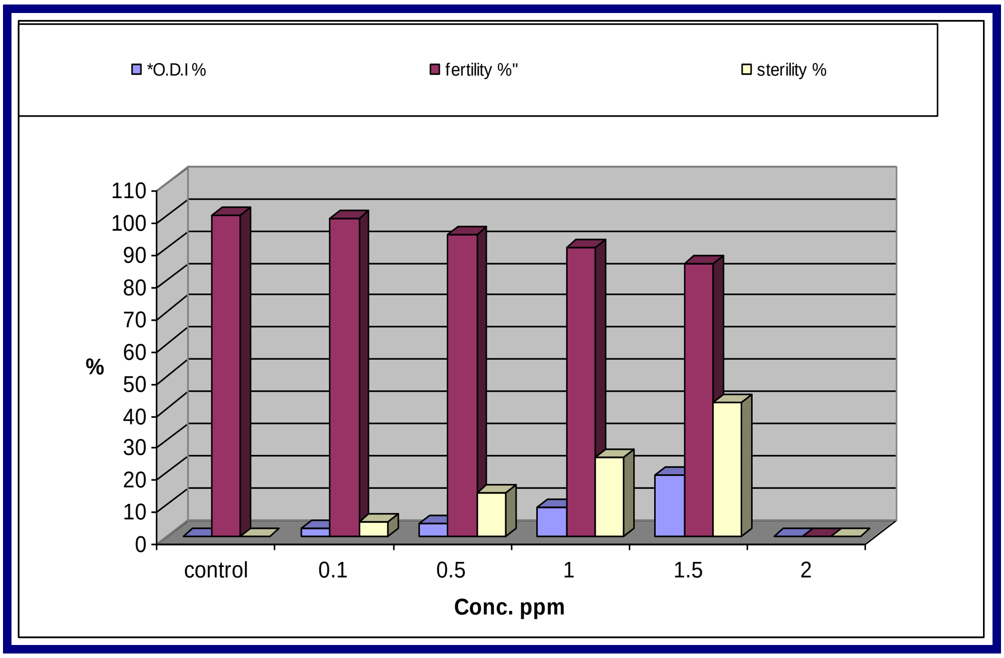


Fig. (9): Effect of Match on (O.D.I , fertility and sterility) of *S. littoralis* treated as 2nd larval instar.

4.1.4. Effect of Match on some biological aspects of *S. littoralis* treated as 4th larval instar:

The data present in **Table (7)** and **Fig. (10)** showed that the corrected percentages of larval mortality had a positive relationship with the different concentrations of Match The percentages of larval mortality were 25, 60, 80, 92.5 and 100 % at the concentrations of 1, 3, 5, 7 and 9 ppm, respectively. The pupation percentages were significantly reduced to 75, 40, 20, 7.5 and 0.0 % as compared with control, while pupal mortality was increased with the increase in concentrations.

Percentage of the adult emergence was decreased significantly with the increase in the concentrations as indicated by 70, 26.25, 3.75 and 2.5 % as compared with control at ascending successive concentrations. On the other hand the present results indicated that there was a significant inhibition of adult emergence percentages as compared with control at the above concentrations.

It is remarkable that the larval and pupal durations were increased with the increasing of concentrations as compared with control, (i.e. the higher concentration induce more prolongation in both larval and pupal durations), as indicated in **Table (7)** and **Fig. (11)**.

Results obtained in **Table (8)** and represented in **Fig. (12)** declared that there was an significantly reduction in both fecundity and fertility, on the contrast (O.D.I) and sterility showed an positive relationship with the increasing of concentrations.

Table (7): The effect of Match on some biological aspects of the cotton leaf worm by feeding newly 4th instar larvae on treated Castor Leaves for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0	6 ±0.41	100 ±0	----- ±0	7 ±0.41	100 ±0	----- ±0
1	***25 ±0.41	7 ±0.41	***75 ±0.41	5 ±0	8 ±0.41	***70 ±0.41	***30 ±2.04
3	***60 ±0.41	*8 ±0.41	***40 ±0.41	***13.75 ±0.29	***10 ±0.41	***26.25 ±0.25	***73.75 ±1.25
5	***80 ±0.58	***9 ±0.41	***20 ±0.41	***16.25 ±0.75	***10 ±0.41	***3.75 ±0.25	***96.25 ±1.25
7	***92.5 ±0.29	***11 ±0	***7.5 ±0.29	5 ±0	***11 ±0.58	***2.5 ±0.29	***97.5 ±1.44
9	***100 ±0	***3 ±0.41	***----- ±0	----- ±0	***----- ±0	***----- ±0	***100 ±0

Table (8): Effect of Match on fecundity, fertility and sterility against adults of cotton leaf worm emerged from 4th larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	$^{+}$O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1340 ±21.6	------ ±0	100 ±0	------ ±0
1	***1100 ±35.35	***9.89 ±0.96	90.77 ±3.38	***25.43 ±3.83
3	***1100 ±35.35	***11.13 ±0.57	***79.58 ±0.88	***34.63 ±2.39
5	***1155 ±20.72	***7.42 ±0.37	***73.49 ±3.39	***36.68 ±2.73
7	***950 ±20.41	***17.04 ±0.41	***57.84 ±3.11	***59.01 ±2.13
9	***------- ±0	------- ±0	***------- ±0	------- ±0

Significant at P = 0.05** * High significant at P = 0.01**

***** Very high significant at P = 0.001** **+ Oviposition deterrenet index**

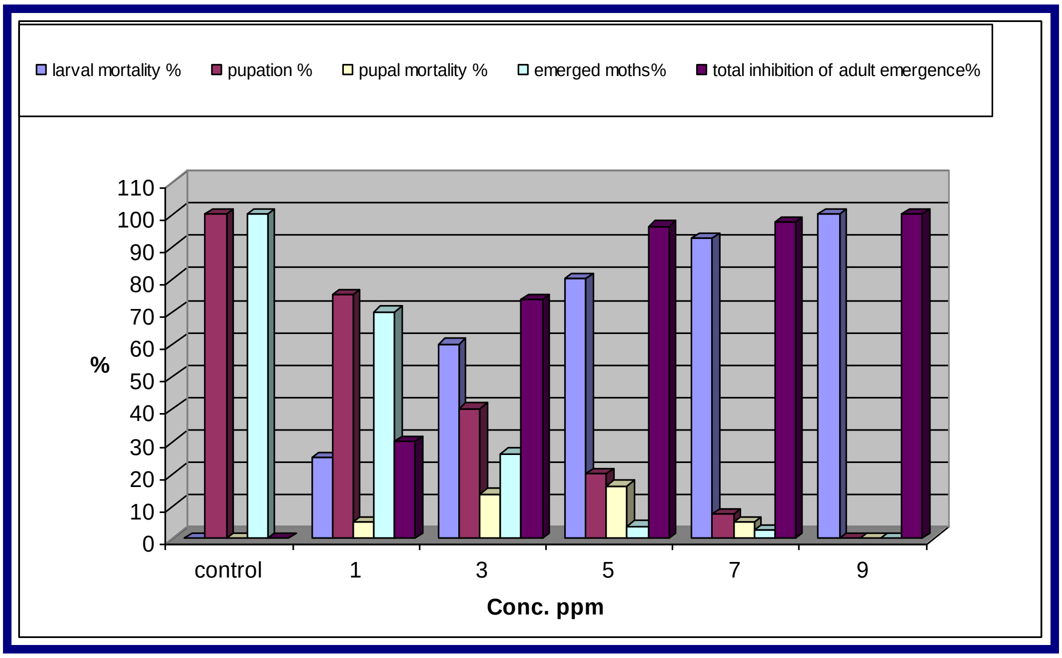


Fig. (10): Effect of Match on some biological aspects of

S. littoralis treated as 4th larval instar.

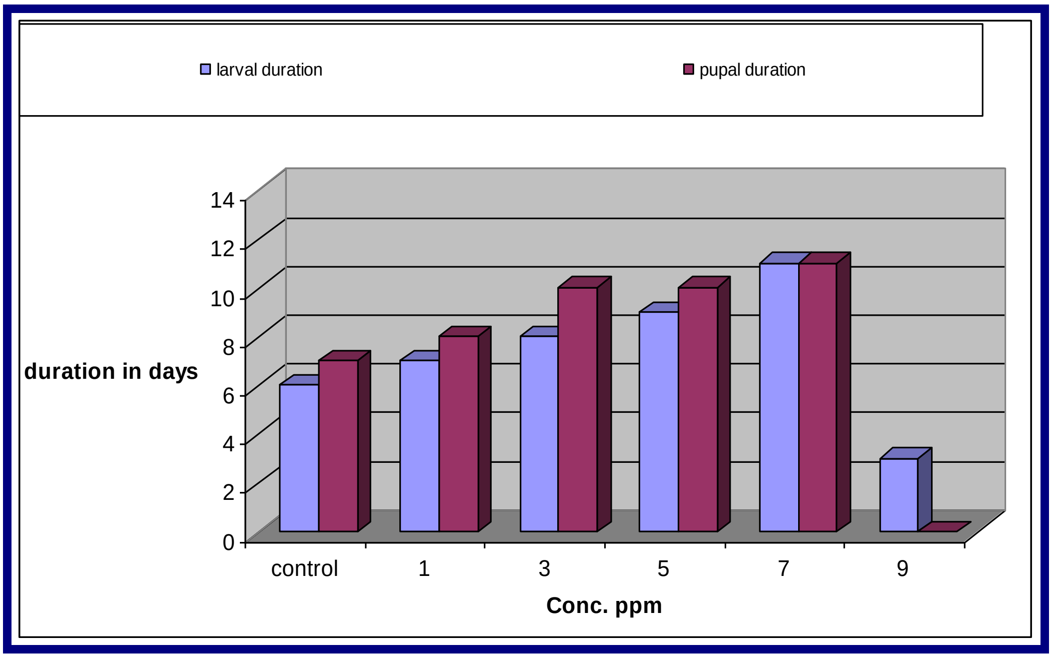


Fig. (11): Effect of Match on larval and pupal duration

of *S. littoralis* treated as 4th larval instar.

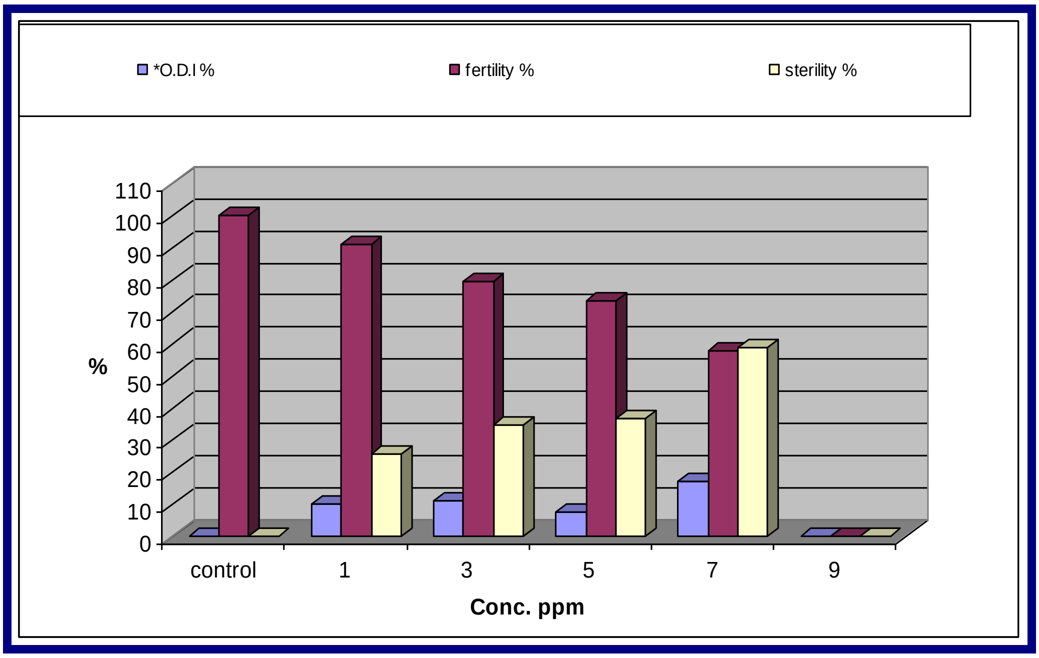


Fig. (12): Effect of Match on (O.D.I , fertility and sterility) of *S. littoralis* treated as 4th larval instar.

4.1.5. Effect of Mimic on some biological aspects of *S. littoralis* treated as 2nd larval instar:

Results recorded in **Table (9)** and **Fig. (13)** declared that there was a significant effect on the larval mortalities that given in corrected percentages by 20, 45, 75, 85 and 100 % at the concentrations of 1, 1.5, 2, 2.5 and 3 ppm, respectively. Also, the pupation percentages were greatly reduced to 80, 55, 25, 15 % as compared with 100 % in control, on the other hand percentages of pupal mortality was increased with the increase in concentrations

The percentage of the adult emergence was decreased with an increase in the concentrations as indicated by 60, 25, 5 and 0.0 % respectively, at ascending successive concentrations. The present results indicated that there was a significant inhibition of adult emergence percentages as 40, 75, 95 and 100 % as compared with control 0.0 %. Larval and pupal durations were increased as shown in **Table (9)** and **Fig. (14)**.

Results obtained in **Table (10)** and illustrated in **Fig. (15)** declared that there was a reduction in both fecundity and fertility, on the contrast (O.D.I) and sterility showed an positive relationship with the increasing of concentrations.

Table (9): The effect of Mimic on some biological aspects of the cotton leaf worm by feeding newly 2nd instar larvae on treated Castor Leaves for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0	10 ±0.41	100 ±0	----- ±0	7 ±0.29	100 ±0	----- ±0
1	*** 20 ±0.41	** 12 ±0.41	*** 80 ±0.41	*** 20 ±0.41	* 9 ±0.41	*** 60 ±0	*** 40 ±0
1.5	*** 45 ±0.41	*** 13 ±0	*** 55 ±0.41	*** 30 ±0.48	*** 10 ±0.41	*** 25 ±0.82	*** 75 ±4.08
2	*** 75 ±0	*** 13 ±0.41	*** 25 ±0	*** 20 ±0.41	*** 11 ±0.41	*** 5 ±0.41	*** 95 ±2.04
2.5	*** 85 ±0.41	*** 14 ±0	*** 15 ±0.41	*** 15 ±0.41	*** 12 ±0	*** ----- ±0	*** 100 ±0
3	*** 100 ±0	*** 5 ±0.41	*** ----- ±0	----- ±0	*** ----- ±0	*** ----- ±0	*** 100 ±0

Table (10): Effect of Mimic on fecundity, fertility and sterility against adults of cotton leaf worm emerged from 2nd larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	+O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1450 ±28.9	------ ±0	100 ±0	------ ±0
1	1420 ±30.34	1.04 ±0.09	** 95.8 ±0.43	* 6.19 ±0.27
1.5	*1355 ±19.5	*** 3.37 ±0.99	*** 92.23 ±1.29	*** 13.7 ±2.6
2	*** 1260 ±14.14	*** 6.84 ±0.5	*** 89.27 ±0.39	*** 22.4 ±0.9
2.5	*** ------ ±0	------ ±0	*** ------ ±0	------ ±0
3	*** ------ ±0	------ ±0	*** ------ ±0	------ ±0

Significant at P = 0.05** * High significant at P = 0.01**

***** Very high significant at P = 0.001** **+ Oviposition deterrenet index**

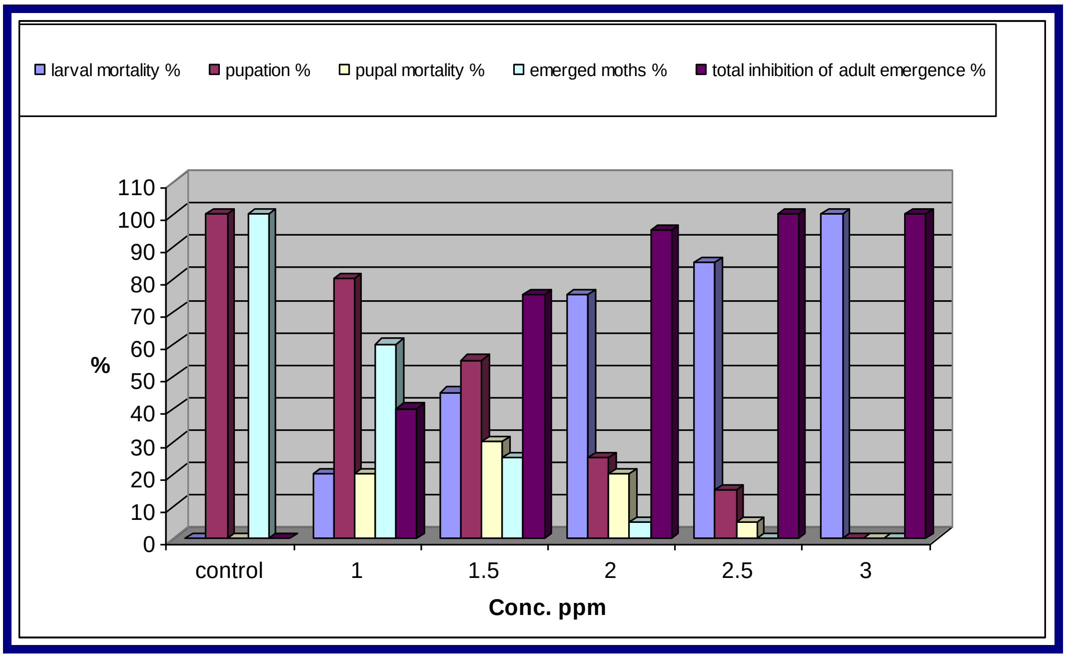


Fig. (13): Effect of Mimic on some biological aspects of

S. littoralis treated as 2nd larval instar.

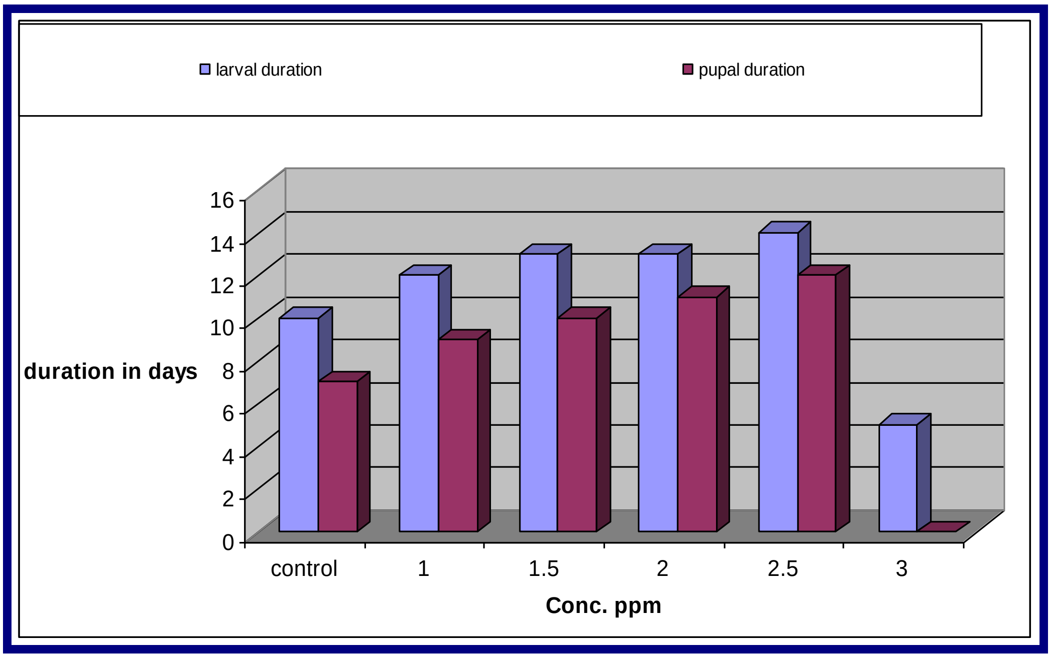


Fig. (14): Effect of Mimic on larval and pupal duration

of *S. littoralis* treated as 2nd larval instar.

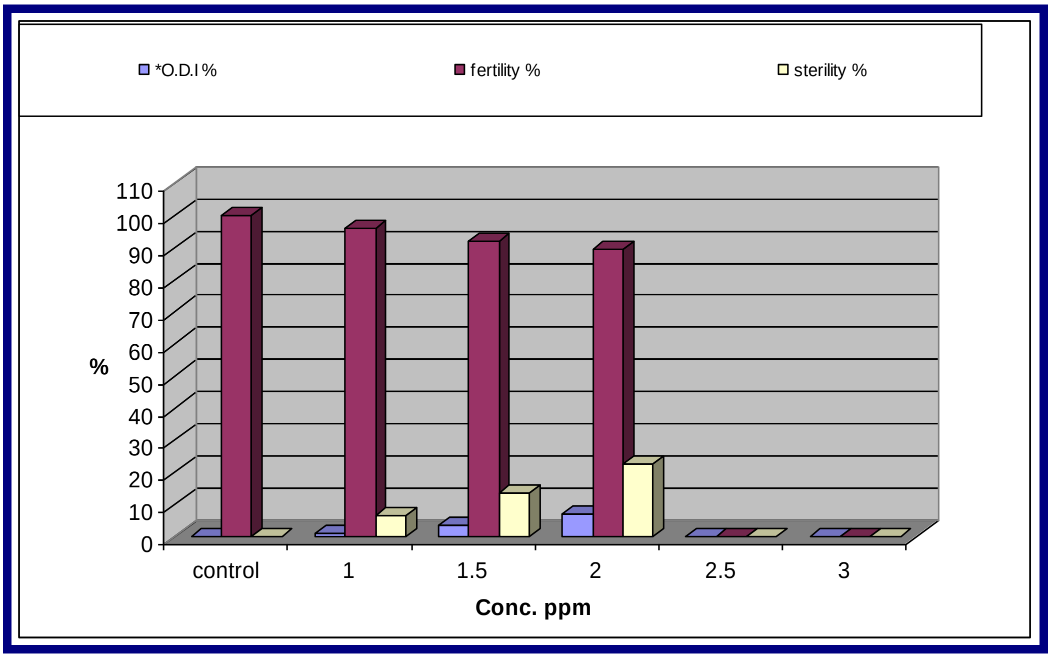


Fig. (15): Effect of Mimic on (O.D.I , fertility and sterility) of *S. littoralis* treated as 2nd larval instar.

4.1.6. Effect of Mimic on some biological aspects of *S. littoralis* treated as 4th larval instar:

Data which tabulated in **Table (11)** and **Fig. (16)** showed that the corrected percentages of larval mortality had a positive relationship with the different concentrations of Mimic, The percentages of larval mortality were 20, 35, 70, 85 and 100 % at the concentrations of 5, 8, 11, 14 and 17 ppm, respectively. Pupation percentages were greatly reduced to 80, 65, 30, 15 and 0.0 % at the above concentrations as compared with control, while pupal mortality was increased with the increase in concentrations.

Percentage of the adult emergence was decreased with an increase in the concentrations as indicated by 70, 50, 11.25 and 2.5 % as compared with control at ascending successive concentrations. On the other hand the results indicated that there was a significant inhibition of adult emergence percentages as compared with control at the above concentrations.

Results in **Table (11)** and **Fig. (17)** showed that the larval and pupal durations were increased with the increasing of concentrations as compared with control, (i.e. the higher concentration induce more prolongation in both larval and pupal durations).

Results obtained in **Table (12)** and illustrated in **Fig. (18)** declared that there was a significant reduction in both fecundity and fertility, on the contrast (O.D.I) and sterility showed an positive relationship with the increasing of concentrations.

Table (11): The effect of Mimic on some biological aspects of the cottonleaf worm by feeding newly 4th instar larvae on treated Castor Leaves for 24hrs.

Conc. (ppm)	Larval mortality % ±S.E	Larval duration (days) ±S.E	Pupation % ±S.E	Pupal mortality % ±S.E	Pupal duration (days) ±S.E.	Emerged moths % ±S.E	Total inhibition of adult emergence %
0.0	----- ±0.0	6 ±0.0	100 ±0.0	----- ±0.0	7 ±0.0	100 ±0.0	----- ±0.0
5	*** 20 ±0.41	*** 8 ±0.0	*** 80 ±0.41	*** 10 ±0.0	** 9 ±0.41	*** 70 ±0.41	*** 30 ±2.04
8	*** 35 ±0.0	*** 8 ±0.41	*** 65 ±0	*** 15 ±0.0	** 9 ±0.41	*** 50 ±0.0	*** 50 ±0.0
11	*** 70 ±0.41	*** 10 ±0.41	*** 30 ±0.41	*** 18.75 ±0.25	*** 10 ±0.41	*** 11.25 ±0.25	*** 88.75 ±1.25
14	*** 85 ±0.41	*** 12 ±0.0	*** 15 ±0.41	*** 12.5 ±0.29	*** 11 ±0.0	*** 2.5 ±0.29	*** 97.5 ±1.25
17	*** 100 ±0.0	*** 3 ±0.0	*** ----- ±0.0	----- ±0.0	*** ----- ±0.0	*** ----- ±0.0	*** 100 ±0.0

Table (12): Effect of Mimic on fecundity, fertility and sterility against adults of cotton leafworm emerged from 4th larval instar feeding on treated castor leaves.

Conc. (ppm)	No. of eggs/female (fecundity) ±S.E	+O.D.I % ±S.E	Egg hatching (fertility) % ±S.E	Sterility % ±S.E
0.0	1375 ±27	------ ±0.0	100 ±0.0	------ ±0.0
5	**1250 ±22.82	*** 4.75 ±0.5	94.4 ±1.5	*** 14.14 ±1.02
8	*** 1145 ±15.54	***9.11 ±0.4	90.4 ±0.6	*** 24.7 ±0.9
11	***1095 ±13.23	*** 11.32 ±0.5	*** 82.06 ±4.4	*** 34.7 ±3.28
14	*** 980 ±31.36	*** 16.81 ±1.2	*** 77.44 ±1.5	*** 44.74 ±2.34
17	*** ------- ±0.0	------- ±0.0	*** ------- ±0.0	------- ±0.0

Significant at P = 0.05** * High significant at P = 0.01**

***** Very high significant at P = 0.001** **+ Oviposition deterrenet index**

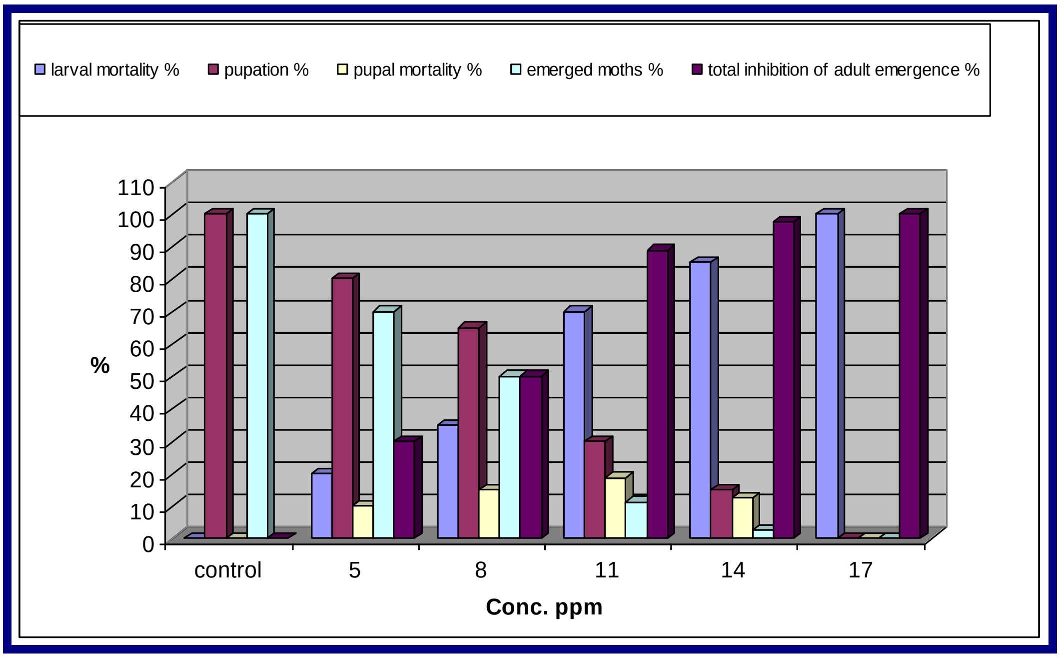


Fig. (16): Effect of Mimic on some biological aspects of *S. littoralis* treated as 4th larval instar.

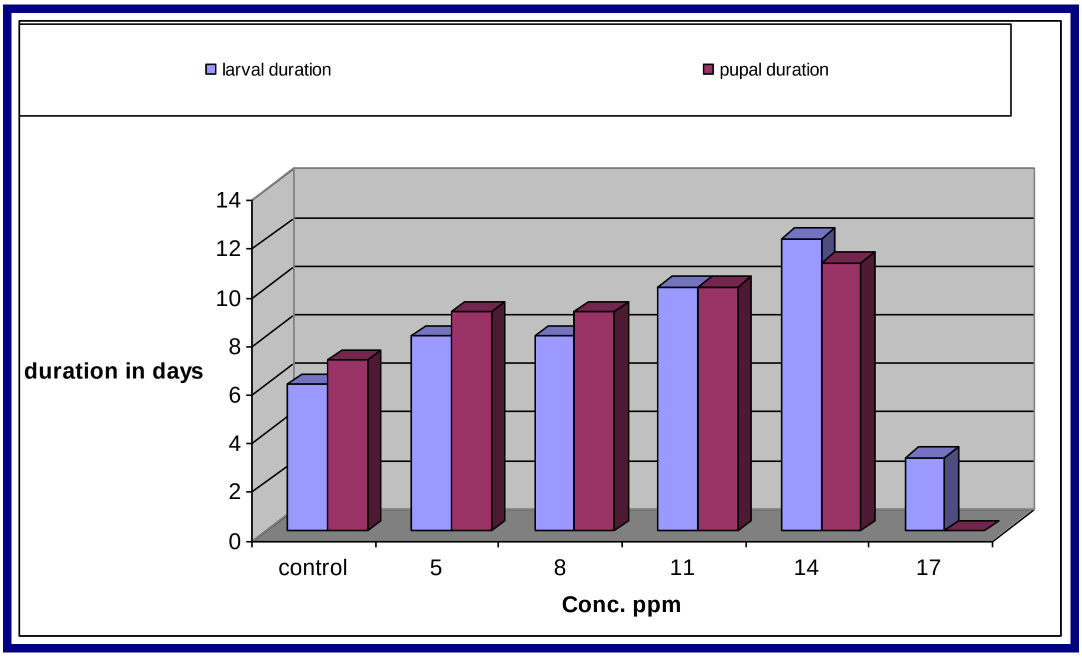


Fig. (17): Effect of Mimic on larval and pupal duration of *S. littoralis* treated as 4th larval instar.

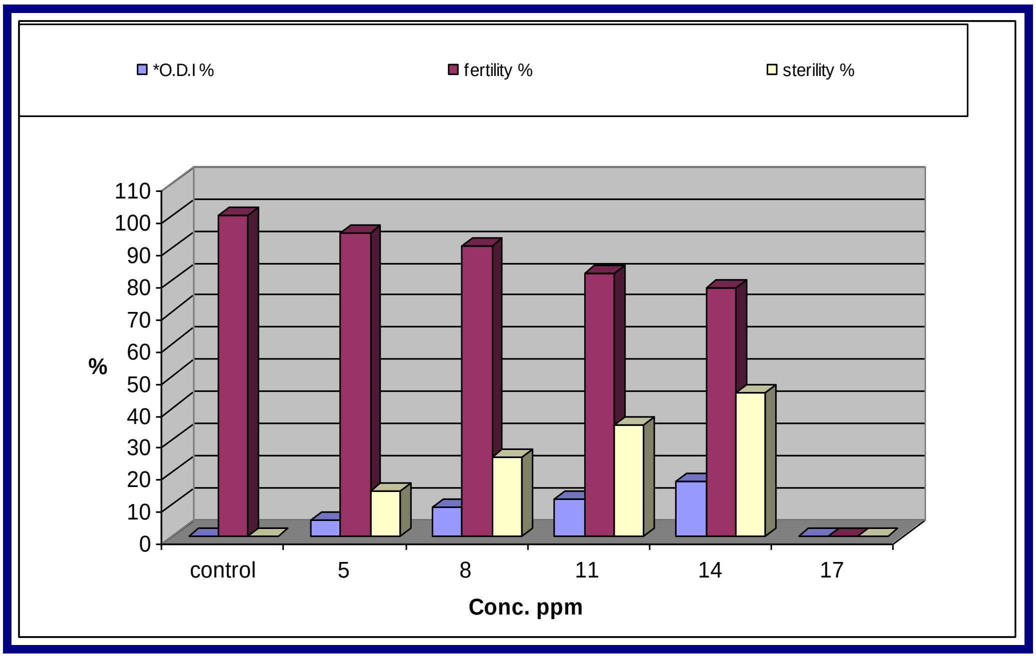


Fig. (18): Effect of Mimic on (O.D.I , fertility and sterility) of *S. littoralis* treated as 4th larval instar.

4.2- Toxicological studies and oral toxicity of the tested insect growth regulators against 2nd and 4th larval instars of *S. littoralis*:-

4.2.1 Susceptibility of 2nd larval instar:

Toxicological activity of the tested IGRs are summarized in **Table (13)**, the tested IGRs could be arranged descendingly according to their potency against 2nd larval instar of *S. littoralis* at the LC_{50} level as following: Cascade, Match and Mimic. The corresponding concentrations (LC_{50}) were: 0.2, 0.6 and 1.5 ppm. respectively, whereas the toxicity lines are drawn in **Fig. (19) and Fig. (20)**. Cascade exhibited the highest toxic action against 2nd larval instar followed by Match while Mimic was the least in toxicity. Obtained results in **Table (13)** showed that the toxicity index at LC_{50} level was as follows: Match (33.33%) and Mimic (13.33%) as toxic as Cascade.

4.2.2 Susceptibility of 4th larval instar:

The tested IGRs could be arranged descendingly according to their potency against 4th larval instar of *S. littoralis* at the LC_{50} level as following: Match, Cascade and Mimic, the corresponding concentrations were: 2, 3.6 and 8.4 ppm respectively, **table (14)**, whereas the toxicity lines are drawn in **Fig. (21)** and **Fig. (22)**. At the LC_{90} level the tested IGRs could be arranged as the same at LC_{50} level and the corresponding concentrations were: 6.3, 9.6 and 15 ppm **table (14)**. Obtained results in **Table (14)** showed that the toxicity index at LC_{50} level was as follows: Cascade (55.55%) and Mimic (23.81%) as toxic as Match, and atLC_{90} level Cascade (64.3%) and Mimic (42%) as toxic as Match.

Table (13) Toxicity data of the tested IGRs against 2nd larval instar of *S. littoralis*.

IGRs	2nd instar				
Conc. (ppm)	LC_{50}	LC_{90}	Slop function	Toxicity index	
				LC_{50}	LC_{90}
Cascade	0.2	1.3	1.8	100	100
Match	0.6	2.5	2.1	33.33	52
Mimic	1.5	2.5	5.6	13.33	52

Table (14) Toxicity data of the tested IGRs against 4th larval instar of *S. littoralis*.

IGRs	4th instar				
Conc. (ppm)	LC_{50}	LC_{90}	Slop function	Toxicity index	
				LC_{50}	LC_{90}
Match	2	6.3	2.6	100	100
Cascade	3.6	9.8	3	55.55	64.3
Mimic	8.4	15	5	23.81	42

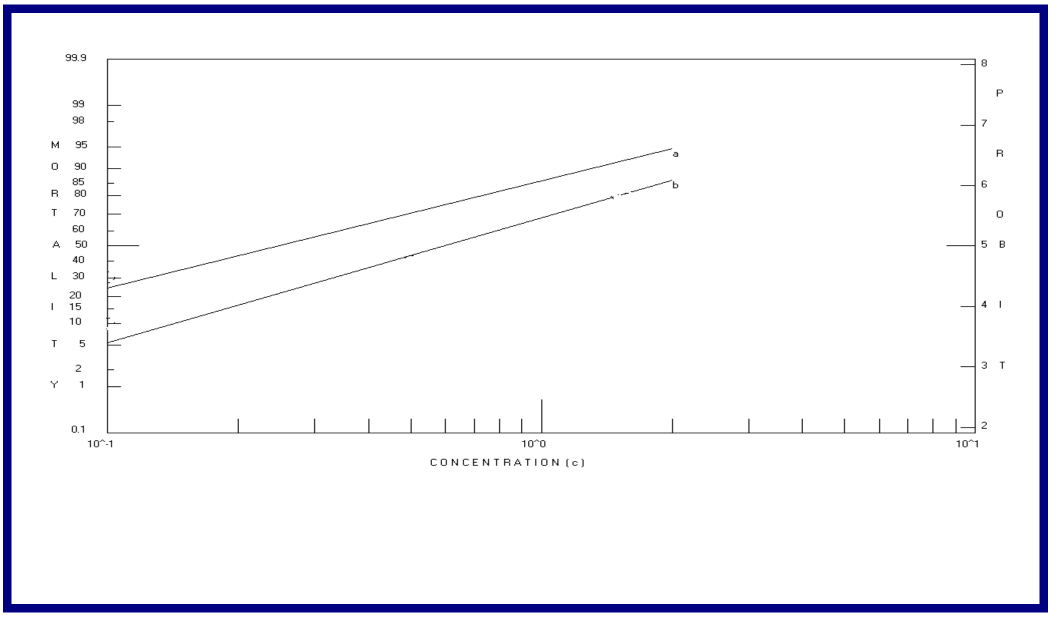


Fig. (19) Regression lines of [Cascade and Match] against 2nd larval instar of *S. littoralis*.

a) Cascade **b) Match**

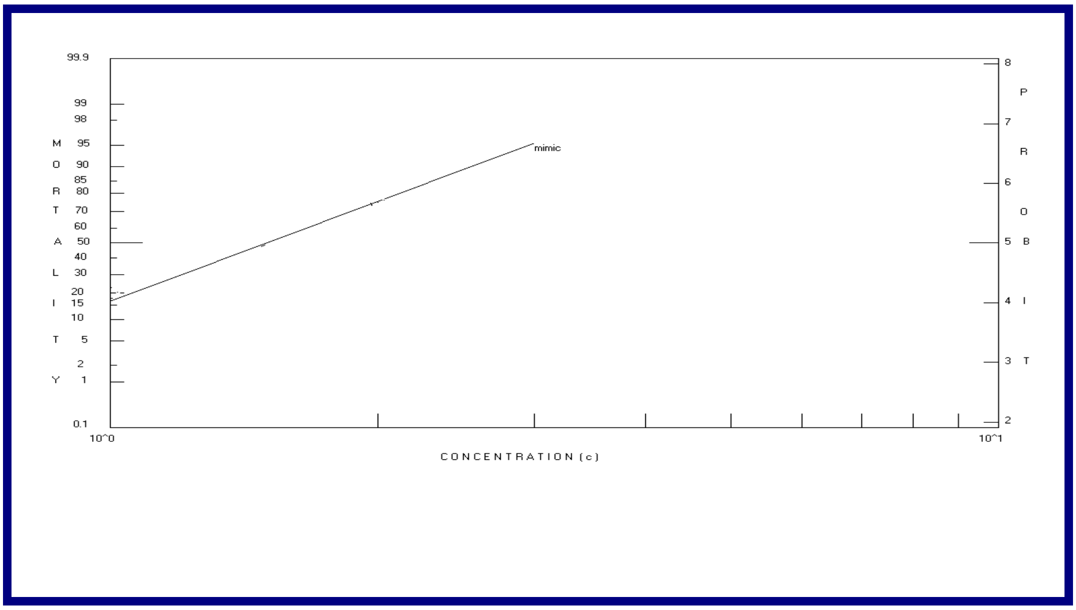


Fig. (20) Regression line of Mimic against 2nd larval instar of *S. littoralis*.

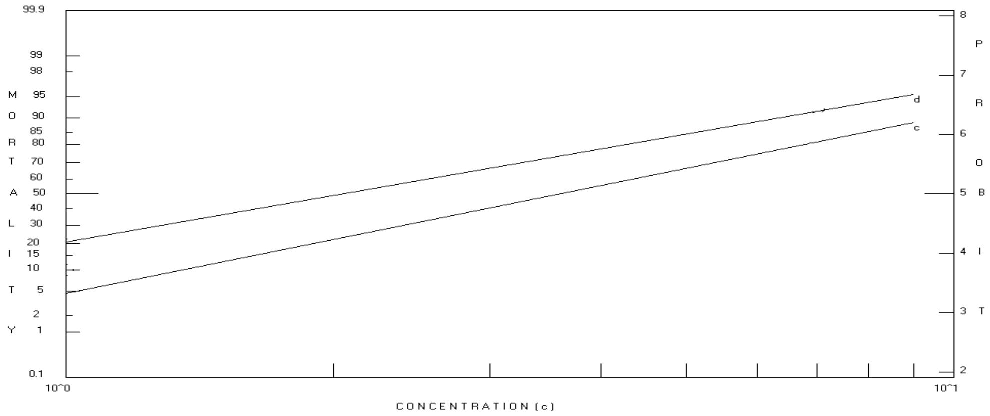


Fig. (21) Regression lines of [Cascade and Match] against 4th larval instar of *S. littoralis.*

c) Cascade d) Match

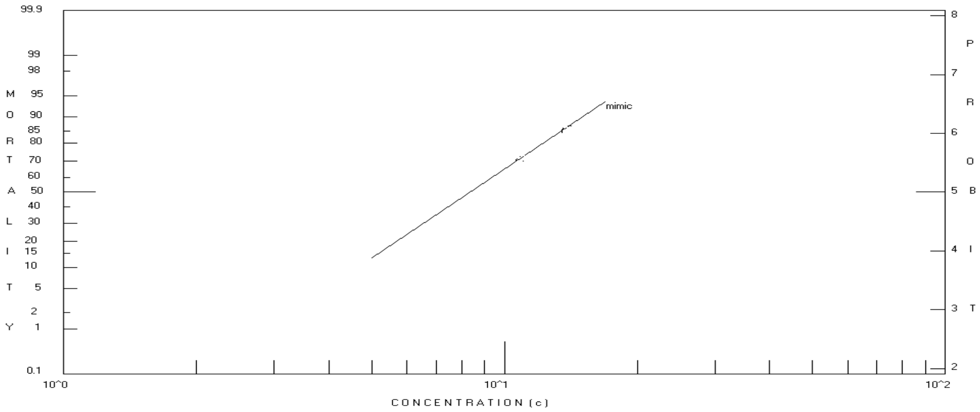


Fig. (22) Regression line of Mimic against 4th larval instar of *S. littoralis.*

4.3-Morphogenic abnormalities and malformation shapes:-

The morphogenic abnormalities of larvae, pupae and adults which emerged from 2nd and 4th larval instars treated with the tested IGRs could be grouped into five categories (malformed 2nd larval instar, malformed 4th larval instar, larval-pupal intermediates, malformed pupae and malformed adults).

4.3.1 Malformed Larvae

As compared with normal 2nd & 4th larval instars, treatments with the different concentrations of the tested IGRs were showed the presence of different degrees of abnormalities in larval stages **Plates (1) & (2)**.

4.3.2 Larval-pupal intermediates:

As compared with normal 2nd larval instar **Plate (1-A)** and with normal 4th larval instar **Plate (2-A)**, the larval-pupal intermediates with different shapes were detected and grouped in **Plate (3)**.

4.3.3 Malformed Pupae:

Treatments of *S. littoralis* larvae in both instars 2nd and 4th with the tested IGRs produced pupae with different degrees of morphogenic abnormalities such as pupa with C- shaped, pupae with a ring of larval cuticle around the abdomen and pupae with enlarged and shortened body **Plate (4)**.

4.3.4 Malformed Adults:

Some emerged adults have various degrees of morphogenic abnormalities. Adults were unable to emerge from their pupal skins (failure adults' emergence), adults were completely free but possessed crumpled and incomplete formation of wings **Plate (5)**.

Plate (1)

(A) **Normal 2nd larval instar (dorsal, lateral and abdominal view).**

(B) **Shrinked 2nd larval instar with greenish chitinization on thorax.**

(C) **3rd larval instar failed to shed the exuvia of 2nd instar.**

(D) **Intermediate instar between 2nd and 3rd instars showed slight chitinization.**

Plate (1)

(A)

(B)

(C)

(D)

Plate (2)

(A) **Normal 4th larval instar**.

(B) **Larva with swollen abdomen with the cuticle color turned to uniform dark gray losing spotted pattern characteristic of the species.**

(C) **Shrinked larva unable to form normal prepupa.**

(D) **Giant larva with dark pigment unable to form normal pre pupa.**

(E), (F) **larvae with malformed thoracic legs and abdominal prolegs.**

(G) **The larva has a swell abdomen, which finally lead to rupture of larva.**

(H), (I) **C-Shaped larva with dark pigments unable to form normal pupa.**

(J) **Intermediate instar between 4th and 5th larval instar with dark pigments unable to molt into 6th instar.**

Plate (2)

(A)

(B) (C) (D) (E) (F)

(G) (H) (I) (J)

Plate (3)

Different shapes of larval-pupal intermediates.

Plate (3)

Plate (4)

(A) **Normal pupa.**

(B) **C- shaped pupa.**

(C), (D) **pupae with large malformed head and small abdomen.**

(E), (F) **Shrinked pupa produced dwarf adult.**

(G) **Small discolored soft-shinned pupa with the rest of larval cuticle around the thorax and head.**

(H) **Pupa with enlarged head and shortened body.**

(I) **Giant pupa with very large malformed head and very small head.**

(J) **Pupa failed in shedding off the larval exuvium**

(K), (L), (M) **Pupa cannot escape from their larval exuvium.**

Plate (4)

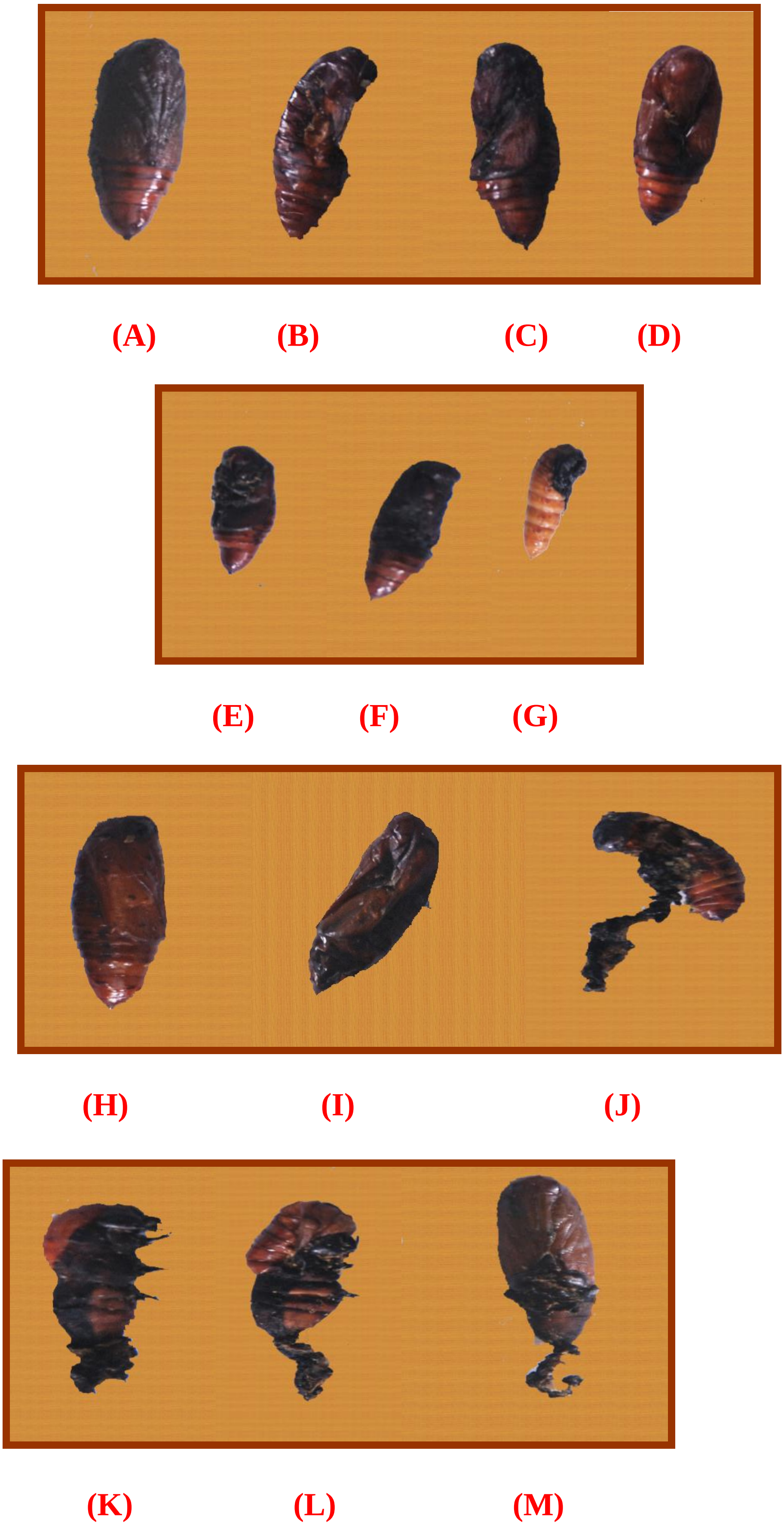


Plate (5)

(A) Normal adult.

(B) Adult has curled wings, which cannot fly.

(C) Adult with malformed abdominal tip, poorly developed curled wings and absence of right antenna.

(D) Poorly developed crumpled wings, malformed legs and abdomen.

(E) Pupal –adult intermediate with head, thoracic legs and wings are free

(F) Adult with poorly developed crumpled twisted wings.

(G) Adult with malformed frizzled wings.

(H) Pupal-adult intermediate with head and thorax enclosed by old cuticle of pupa.

(I) Frizzled wings, absence of legs and malformed abdomen.

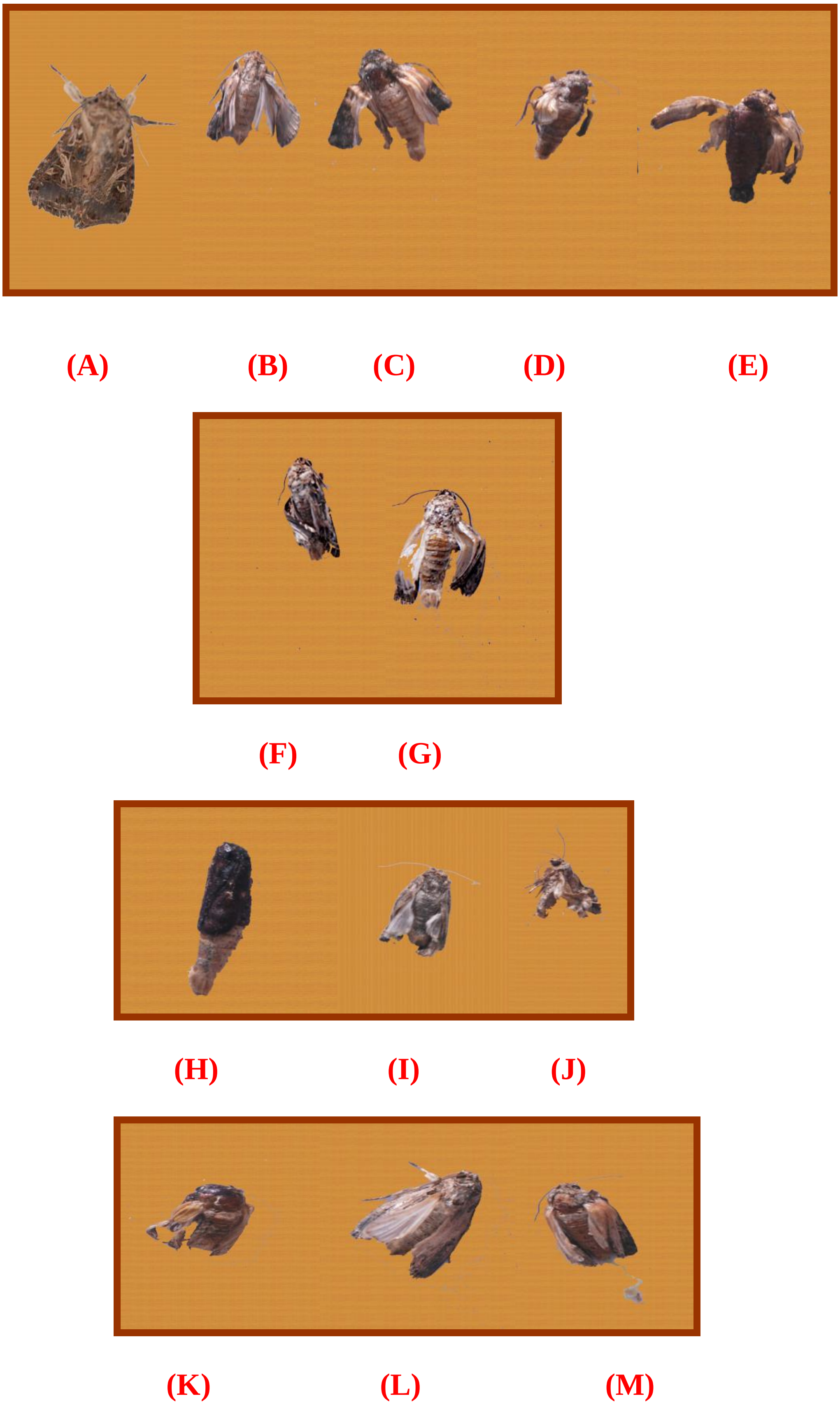
(J) Small adult with twisted wings.

(K) Dwarfed adult with frizzled wings.

(L) Moth with poorly developed crumbled wings.

(M) Adult with twisted wings.

Plate (5)



4.4- Effect of sub- lethal concentrations of the tested IGRs on some enzymatic activities:-

Enzymes were measured in treated and control groups of 2nd and 4th larval instars at 6, 12,24 and 48hrs post treatment with the tested IGRs in order to determine the changes in these enzymes activity through the IGRs mode of action.

4.4.1. Effect of Cascade on acid phosphatase:-

The data recorded in **Table (15) and Fig. (23)** indicated that all the treatments with sub-lethal conc. (LC_{25}, LC_{50} and LC_{90}) on 2nd larval instar at different time intervals have a positive effect, the activities percents were decreased with the increase of time and also with the increase in concentrations.

Also treated 4th larval instar showed a reduction in activities percents with increasing in hours post treatments and in doses as showed in **Table (15)** and **Fig. (24)**.

4.4.2. Effect of Cascade on non-specific esterases (α, β- esterases):-

The results in **Tables (16)&(17)** and **Figs. (25)&(27)** showed that the treatment of 2nd larval instar with sub-lethal doses at different time intervals make an inhibition of activities of both enzymes, with increasing in hours post treatments and in dose.

Data tabulated in **Tables (16)&(17)** and **Figs**. **(26)&(28)** declared that, the activities of non-specific enzymes of the treated 4th larval instars were decreased as hours post treatments and dose were increased.

Table (15): Acid phosphatase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Cascade at different time intervals.

Larval stage	Dose (ppm)	Acid phosphatase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC$_{25}$ (0.1)	6	2.0 ±0.01	1.34 ±0.03 **	-33
		12	9.445 ±0.2	5.23 ±0.1 **	-44.63
		24	11.86 ±0.24	6.23 ±0.31 **	-47.47
		48	13.47 ±0.41	5.93 ±0.34 **	-55.98
	LC$_{50}$ (0.2)	6	2.0 ±0.01	1.0 ±0.1 **	-50
		12	9.445 ±0.2	3.68 ±0.32 **	-61.04
		24	11.86 ±0.24	4.98 ±0.31 **	-58.01
		48	13.47 ±0.41	4.93 ±0.34 **	-63.4
	LC$_{90}$ (1.3)	6	2.0 ±0.01	0.8 ±0.001 **	-60
		12	9.445 ±0.2	2.96 ±0.21 **	-68.66
		24	11.86 ±0.24	3.89 ±0.34 **	-67.2
		48	13.47 ±0.41	3.98 ±0.55 **	-70.45
4th larval instar	LC$_{25}$ (2.1)	6	5.979 ±0.05	5.39 ±0.1 **	-9.85
		12	22.89 ±0.38	18.63 ±0.47 **	-18.61
		24	23.73 ±0.42	16.68 ±0.58 **	-29.71
		48	26.94 ±0.53	18.53 ±0.62 **	-31.22
	LC$_{50}$ (3.6)	6	5.979 ±0.05	4.16 ±0.38 **	-30.42
		12	22.89 ±0.38	16.93 ±0.21 **	-26.04
		24	23.73 ±0.42	15.85 ±0.71 **	-33.21
		48	26.94 ±0.53	16.38 ±0.62 **	-38.085
	LC$_{90}$ (9.8)	6	5.979 ±0.05	3.12 ±0.11 **	-47.82
		12	22.89 ±0.38	14.71 ±0.33 **	-35.74
		24	23.73 ±0.42	12.92 ±0.51 **	-45.55
		48	26.94 ±0.53	13.24 ±0.42 **	-50.85

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

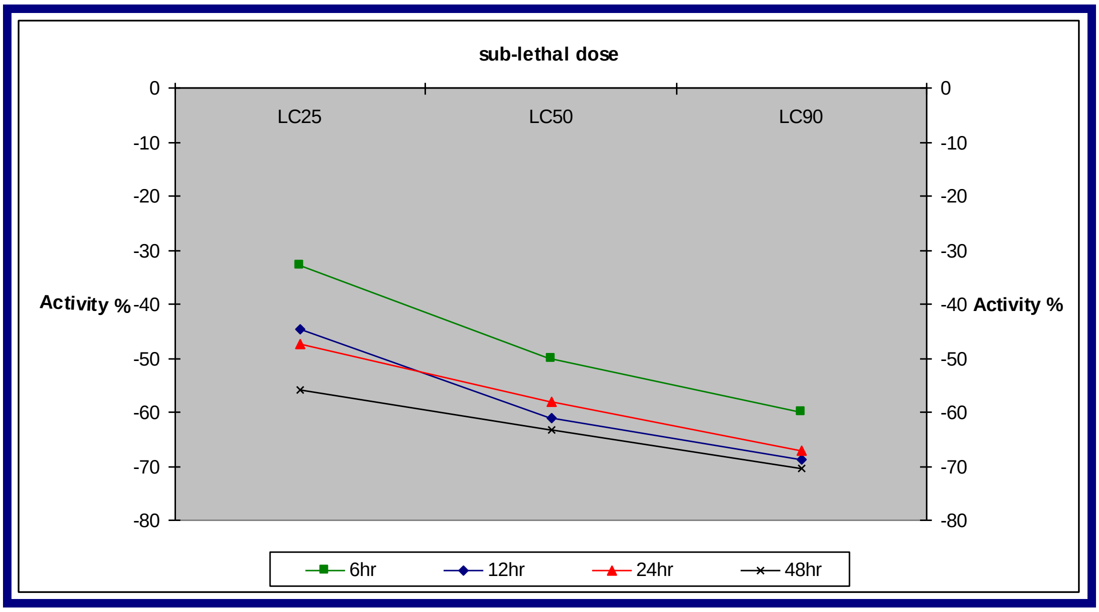


Fig. (23): Acid phosphatase activity of 2nd larval instar treated with Sub-lethal doses of Cascade at different time intervals.

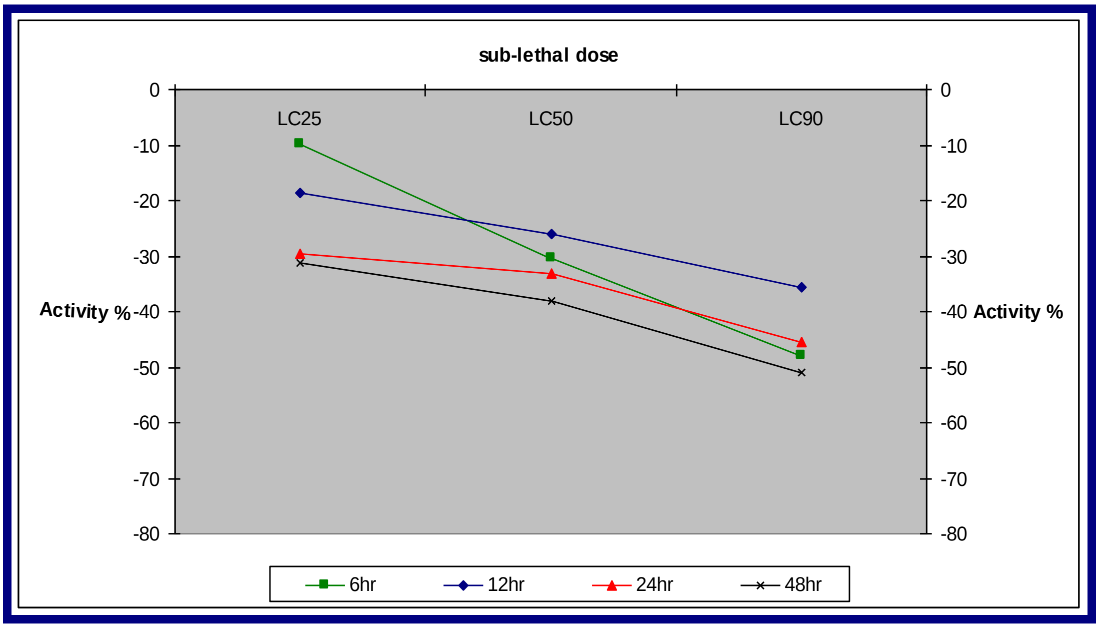


Fig. (24): Acid phosphatase activity of 4th larval instar treated with Sub-lethal doses of Cascade at different time intervals.

Table (16): α-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Cascade at different time intervals.

Larval stage	Dose (ppm)	α-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC$_{25}$ (0.1)	6	464.835 ±2.64	355.835 ±1.34**	-23.45
		12	474.665 ±1.9	347.142 ±2.1**	-26.86
		24	553.33 ±2.4	397.69 ±2.4**	-28.13
		48	673.335 ±3.4	452.76 ±3.1**	-32.76
	LC$_{50}$ (0.2)	6	464.835 ±2.64	339.246 ±4.2**	-27.02
		12	474.665 ±1.9	317.358 ±3.7**	-33.14
		24	553.33 ±2.4	350.269 ±3.2**	-36.7
		48	673.335 ±3.4	413.634 ±2.7**	-38.57
	LC$_{90}$ (1.3)	6	464.835 ±2.64	297.359 ±1.4**	-36.03
		12	474.665 ±1.9	295.478 ±3.7**	-37.75
		24	553.33 ±2.4	306.943 ±1.3**	-44.53
		48	673.335 ±3.4	398.864 ±2.3*	-40.8
4th larval instar	LC$_{25}$ (2.1)	6	749.57 ±3.6	650.67 ±2.2**	-13.19
		12	786.43 ±1.8	638.78 ±4.1**	-18.77
		24	899.67 ±2.8	717.33 ±2.4**	-20.27
		48	976.83 ±2.4	753.27 ±1.9**	-22.89
	LC$_{50}$ (3.6)	6	749.57 ±3.6	636.93 ±2.3**	-15.03
		12	786.43 ±1.8	597.96 ±4.6**	-23.96
		24	899.67 ±2.8	651.89 ±1.7**	-27.54
		48	976.83 ±2.4	663.89 ±3.9**	-32.04
	LC$_{90}$ (9.8)	6	749.57 ±3.6	598.97 ±5.2**	-20.09
		12	786.43 ±1.8	559.89 ±4.1**	-28.81
		24	899.67 ±2.8	596.94 ±3.3**	-33.65
		48	976.83 ±2.4	594.79 ±4.1**	-39.11

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

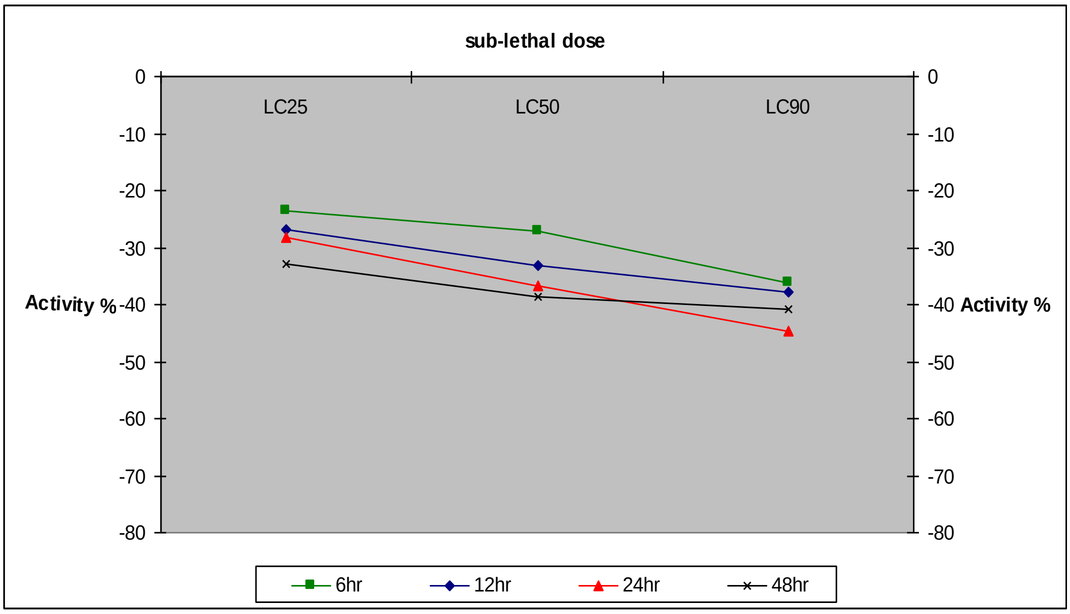


Fig. (25): α-esterase activity of 2nd larval instar treated with Sub-lethal doses of Cascade at different time intervals.

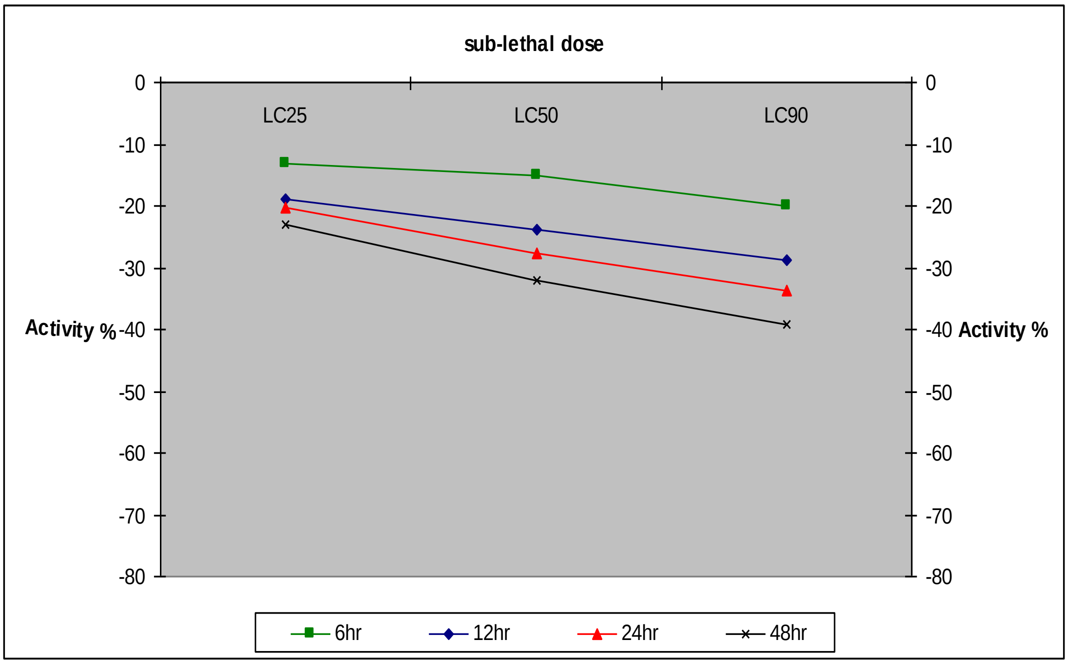


Fig. (26): α-esterase activity of 4th larval instar treated with Sub-lethal doses of Cascade at different time intervals.

Table (17): β-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Cascade at different time intervals.

Larval stage	Dose (ppm)	β-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC$_{25}$ (0.1)	6	619.165 ±5.2	543.5 ±3.7**	-12.22
		12	842.665 ±4.4	700.213 ±3.1**	-16.9
		24	956.5 ±7.6	724.44 ±5.8**	-24.3
		48	1148.33 ±9.3	815.32 ±4.3**	-29.9
	LC$_{50}$ (0.2)	6	619.165 ±5.2	487.33 ±4.6**	-21.3
		12	842.665 ±4.4	620.42 ±6.4**	-26.4
		24	956.5 ±7.6	680.4 ±6.1**	-28.9
		48	1148.33 ±9.3	776.6 ±5.8**	-32.4
	LC$_{90}$ (1.3)	6	619.165 ±5.2	409.22 ±2.2**	-33.91
		12	842.665 ±4.4	533.18 ±7.2**	-36.73
		24	956.5 ±7.6	552.63 ±2.6**	-42.22
		48	1148.33 ±9.3	613.72 ±5.8**	-46.55
4th larval instar	LC$_{25}$ (2.1)	6	1267.33 ±22.3	1153 ±14.2*	-9.02
		12	1734.28 ±32.1	1542.3 ±20.05**	-11.07
		24	1913.42 ±17.4	1596.82 ±15.32**	-16.55
		48	1125.48 ±24.3	1073.6 ±13.5*	-4.09
	LC$_{50}$ (3.6)	6	1267.33 ±22.3	1096.93 ±14.1*	-13.44
		12	1734.28 ±32.1	1406.75 ±18.01**	-18.9
		24	1913.42 ±17.4	1485.74 ±18.4**	-22.35
		48	1125.48 ±24.3	1002.8 ±16.2*	-10.9
	LC$_{90}$ (9.8)	6	1267.33 ±22.3	984.34 ±12.7**	-22.33
		12	1734.28 ±32.1	1300.82 ±21.4**	-25
		24	1913.42 ±17.4	1347.33 ±18.4**	-29.6
		48	1125.48 ±24.3	987.53 ±12.7*	-12.26

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

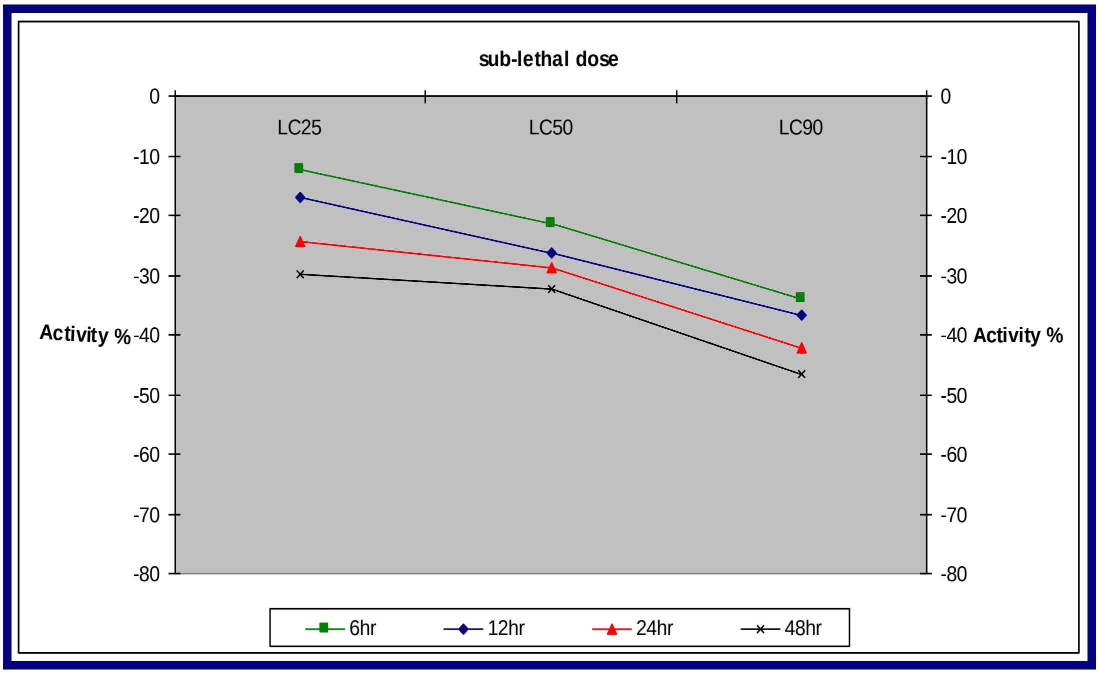


Fig. (27): β-esterase activity of 2nd larval instar treated with Sub-lethal doses of Cascade at different time intervals.

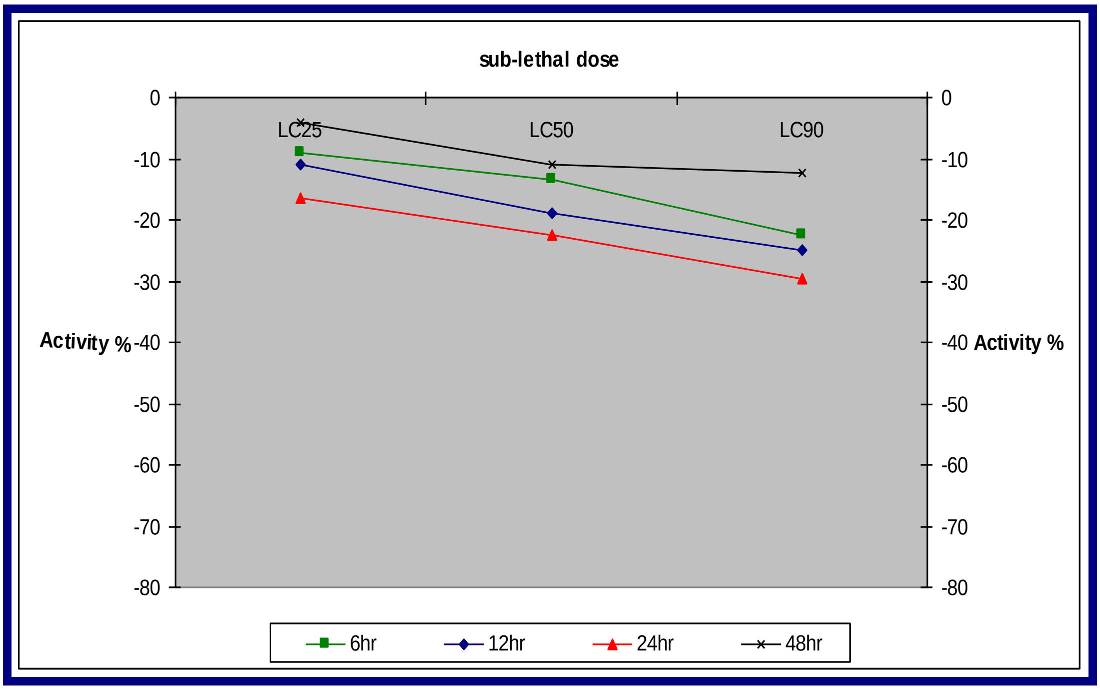


Fig. (28): β-esterase activity of 4th larval instar treated with Sub-lethal doses of Cascade at different time intervals.

4.4.3. Effect of Match on acid phosphatase:-

Results recorded in **Table (18)** and **Fig. (29)** showed that when 2nd larval instar was treated with sub-lethal concentrations the enzymatic activities were inhibited with the increase in the time post-treatment and also with the increase in dose.

Data obtained in **Table (18)** and **Fig. (30)** declared that treated 4th larval instar with sub-lethal concentrations showed a significant decreasing in activity with increasing of time post- treatment and increasing of dose.

4.4.4. Effect of Match on non-specific esterases (α, β- esterases):-

Results obtained and tabulated in **Tables (19)&(20)** and illustrated in **Figures (31)&(33)** declared that treated 2nd larval instar showed an decreasing in enzymatic activities as the dose increase and the time post-treatment increase.

Treated 4th larval instar with sub-lethal doses showed an reduction activities of non-specific esterases as the time post-treatment increase and also with the increasing of the dose, as showed in **Tables (19)&(20)** and illustrated in **Figures (32)&(34)**.

Table (18): Acid phosphatase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Match at different time intervals.

Larval stage	Dose (ppm)	Acid phosphatase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC_{25} (0.3)	6	2.0 ±0.01	1.64 ±0.2 **	-18
		12	9.445 ±0.2	6.1 ±0.24 **	-35.42
		24	11.86 ±0.24	7.64 ±0.38 **	-35.9
		48	13.47 ±0.41	8.43 ±0.34 **	-37.42
	LC_{50} (0.6)	6	2.0 ±0.01	1.1 ±0.05 **	-45
		12	9.445 ±0.2	4.97 ±0.36 **	-47.38
		24	11.86 ±0.24	5.88 ±0.41 **	-50.42
		48	13.47 ±0.41	6.91 ±0.32 **	-48.7
	LC_{90} (2.5)	6	2.0 ±0.01	0.9 ±0.003 **	-55
		12	9.445 ±0.2	3.46 ±0.45 **	-63.37
		24	11.86 ±0.24	4.38 ±0.61 **	-63.16
		48	13.47 ±0.41	5.21 ±0.35 **	-61.32
4th larval instar	LC_{25} (1.1)	6	5.979 ±0.05	4.68 ±0.03 **	-21.73
		12	22.89 ±0.38	16.46 ±0.27 **	-28.1
		24	23.73 ±0.42	14.47 ±0.61 **	-39.022
		48	26.94 ±0.53	15.86 ±0.32 **	-41.13
	LC_{50} (2.0)	6	5.979 ±0.05	3.97 ±0.08 **	-33.6
		12	22.89 ±0.38	15.33 ±0.31 **	-33.03
		24	23.73 ±0.42	13.68 ±0.41 **	-42.35
		48	26.94 ±0.53	14.67 ±0.62 **	-45.54
	LC_{90} (6.3)	6	5.979 ±0.05	2.99 ±0.34 **	-49.99
		12	22.89 ±0.38	10.97 ±0.43 **	-52.07
		24	23.73 ±0.42	10.5 ±0.31 **	-55.75
		48	26.94 ±0.53	11.0 ±0.62 **	-59.17

* Significant at P = 0.05 ** High significant at P = 0.01

*** Very high significant at P = 0.001

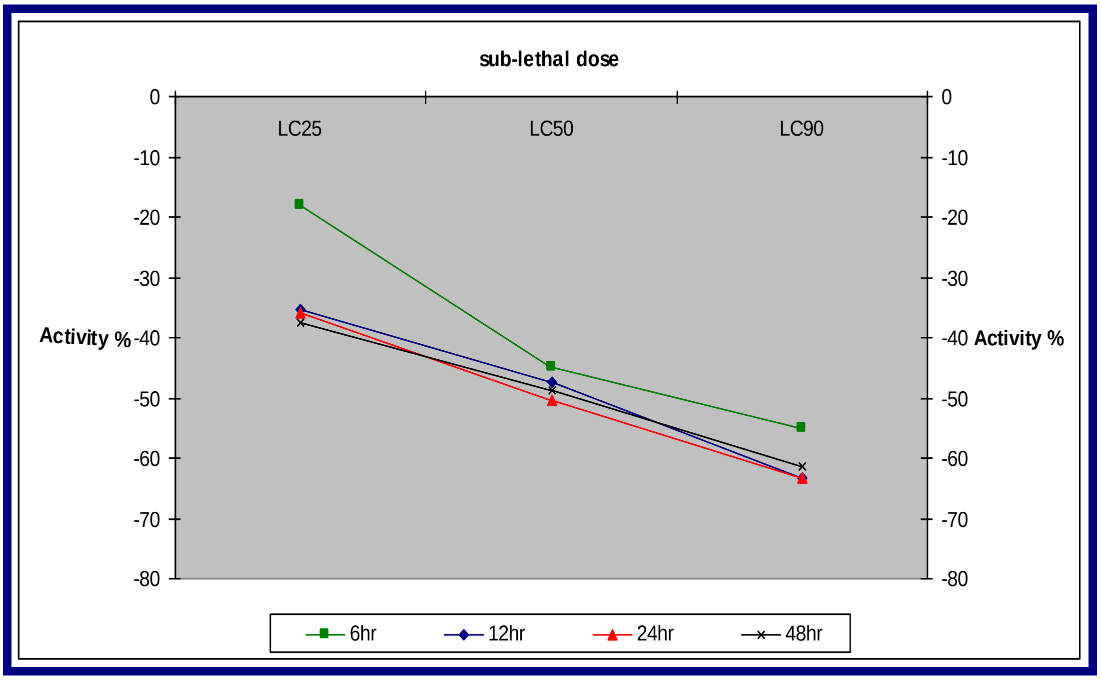


Fig. (29): Acid phosphatase activity of 2nd larval instar treated with Sub-lethal doses of Match at different time intervals.

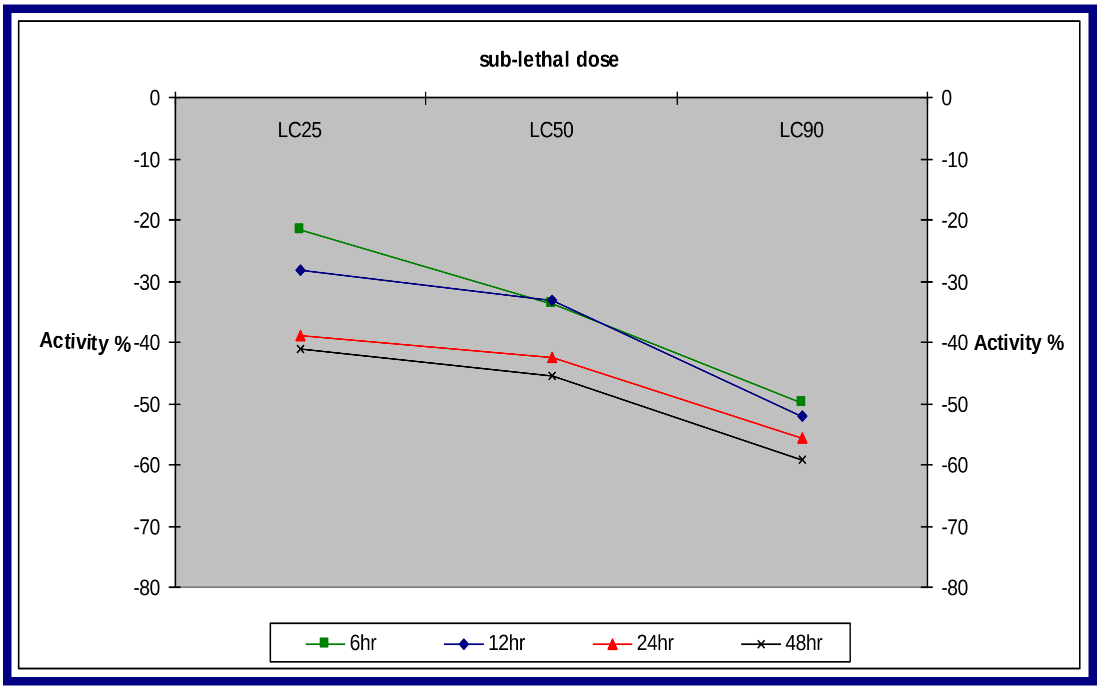


Fig. (30): Acid phosphatase activity of 4th larval instar treated with Sub-lethal doses of Match at different time intervals.

Table (19): α-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Match at different time intervals.

Larval stage	Dose (ppm)	α-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC_{25} (0.3)	6	464.835 ±2.64	370.736 ±3.2*	-20.24
		12	474.665 ±1.9	373.866 ±2.9**	-21.23
		24	553.33 ±2.4	402.936 ±1.6**	-27.18
		48	673.335 ±3.4	498.685 ±3.4*	-25.94
	LC_{50} (0.6)	6	464.835 ±2.64	353.472 ±5.2*	-23.96
		12	474.665 ±1.9	332.647 ±2.6**	-29.92
		24	553.33 ±2.4	400.861 ±3.6**	-27.55
		48	673.335 ±3.4	469.747 ±1.9**	-30.23
	LC_{90} (2.5)	6	464.835 ±2.64	317.189 ±4.5**	-31.76
		12	474.665 ±1.9	299.398 ±5.7**	-36.92
		24	553.33 ±2.4	336.431 ±2.8**	-39.199
		48	673.335 ±3.4	306.597 ±1.4**	-40.06
4th larval instar	LC_{25} (1.1)	6	749.57 ±3.6	598.85 ±3.2**	-20.11
		12	786.43 ±1.8	600.94 ±2.6**	-23.59
		24	899.67 ±2.8	643.83 ±3.3**	-28.44
		48	976.83 ±2.4	686.76 ±4.2**	-29.69
	LC_{50} (2.0)	6	749.57 ±3.6	563.93 ±2.7**	-24.77
		12	786.43 ±1.8	535.96 ±1.8**	-31.85
		24	899.67 ±2.8	548.73 ±4.6**	-39.00
		48	976.83 ±2.4	564.66 ±3.5**	-42.19
	LC_{90} (6.3)	6	749.57 ±3.6	519.73 ±3.3**	-30.66
		12	786.43 ±1.8	514.88 ±2.7**	-34.53
		24	899.67 ±2.8	468.69 ±4.1**	-47.9
		48	976.83 ±2.4	472.56 ±4.6**	-51.62

* Significant at P = 0.05 ** High significant at P = 0.01

*** Very high significant at P = 0.001

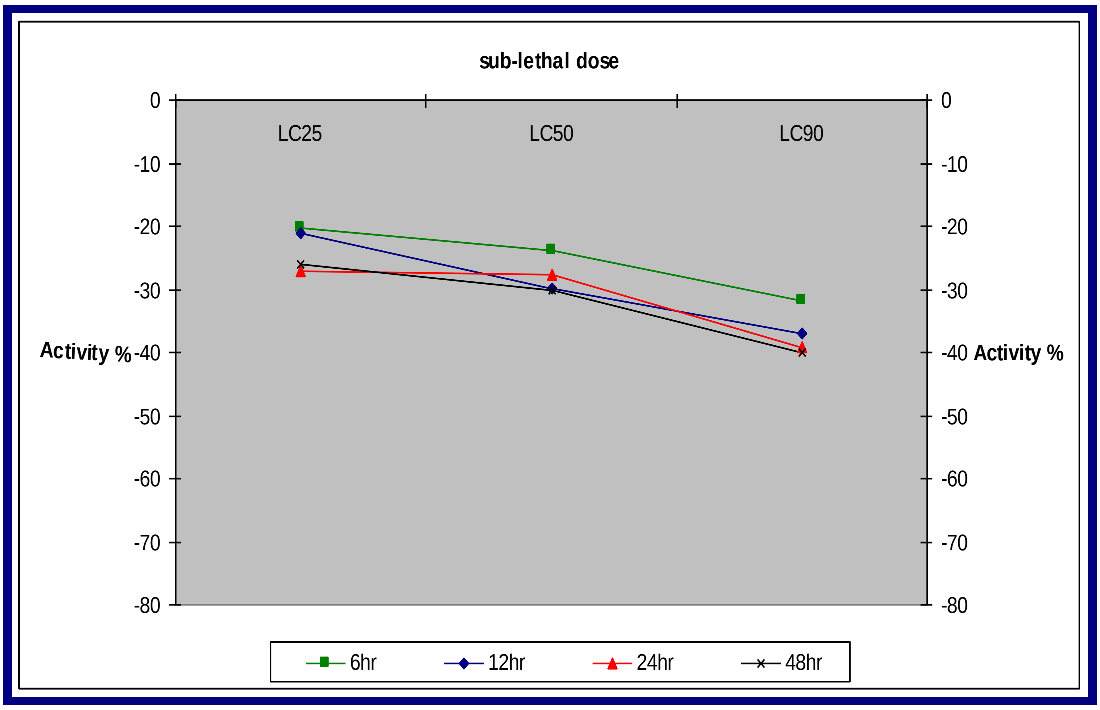


Fig. (31): α-esterase activity of 2nd larval instar treated with Sub-lethal doses of Match at different time intervals.

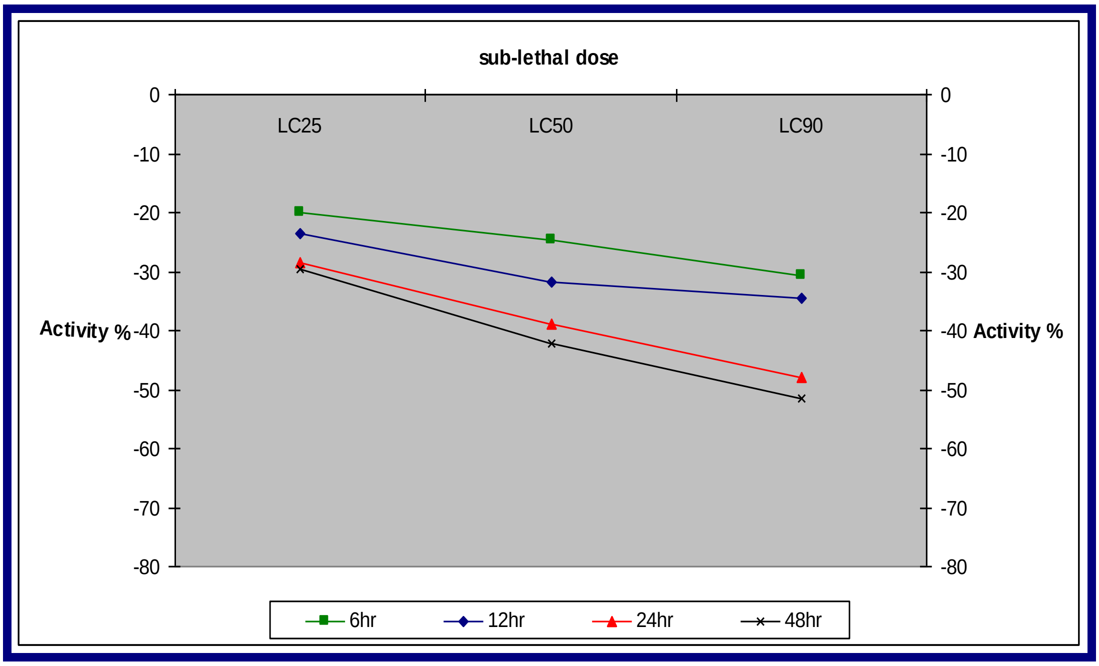


Fig. (32): α-esterase activity of 4th larval instar treated with Sub-lethal doses of Match at different time intervals.

Table (20): β-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Match at different time intervals.

Larval stage	Dose (ppm)	β-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC_{25} (0.3)	6	619.165 ±5.2	560.3 ±8.2*	-9.51
		12	842.665 ±4.4	742.162 ±2.3**	-11.93
		24	956.5 ±7.6	768.36 ±8.9**	-19.67
		48	1148.33 ±9.3	901.11 ±6.4**	-21.53
	LC_{50} (0.6)	6	619.165 ±5.2	525.7 ±11.2**	-15.09
		12	842.665 ±4.4	683.62 ±7.6**	-18.9
		24	956.5 ±7.6	749.52 ±10.4**	-21.64
		48	1148.33 ±9.3	869.41 ±12.8**	-24.29
	LC_{90} (2.5)	6	619.165 ±5.2	475.6 ±9.9**	-23.19
		12	842.665 ±4.4	622.3 ±8.6**	-26.15
		24	956.5 ±7.6	666.14 ±10.7**	-30.36
		48	1148.33 ±9.3	723.9 ±6.8**	-36.96
4th larval instar	LC_{25} (1.1)	6	1267.33 ±22.3	1086.28 ±24.03*	-14.28
		12	1734.28 ±32.1	1423.64 ±26.4**	-17.91
		24	1913.42 ±17.4	1418.54 ±14.21**	-25.9
		48	1125.48 ±24.3	1030 ±12.4*	-8.5
	LC_{50} (2.0)	6	1267.33 ±22.3	980.24 ±20.2**	-22.65
		12	1734.28 ±32.1	1300.73 ±26.03**	-25
		24	1913.42 ±17.4	1328.44 ±15.6**	-30.6
		48	1125.48 ±24.3	978.93 ±19.08**	-13.02
	LC_{90} (6.3)	6	1267.33 ±22.3	882.29 ±11.3**	-30.4
		12	1734.28 ±32.1	1124.14 ±27.6**	-35.2
		24	1913.42 ±17.4	1156.5 ±23.2**	-39.56
		48	1125.48 ±24.3	894.7 ±16.41**	-20.5

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

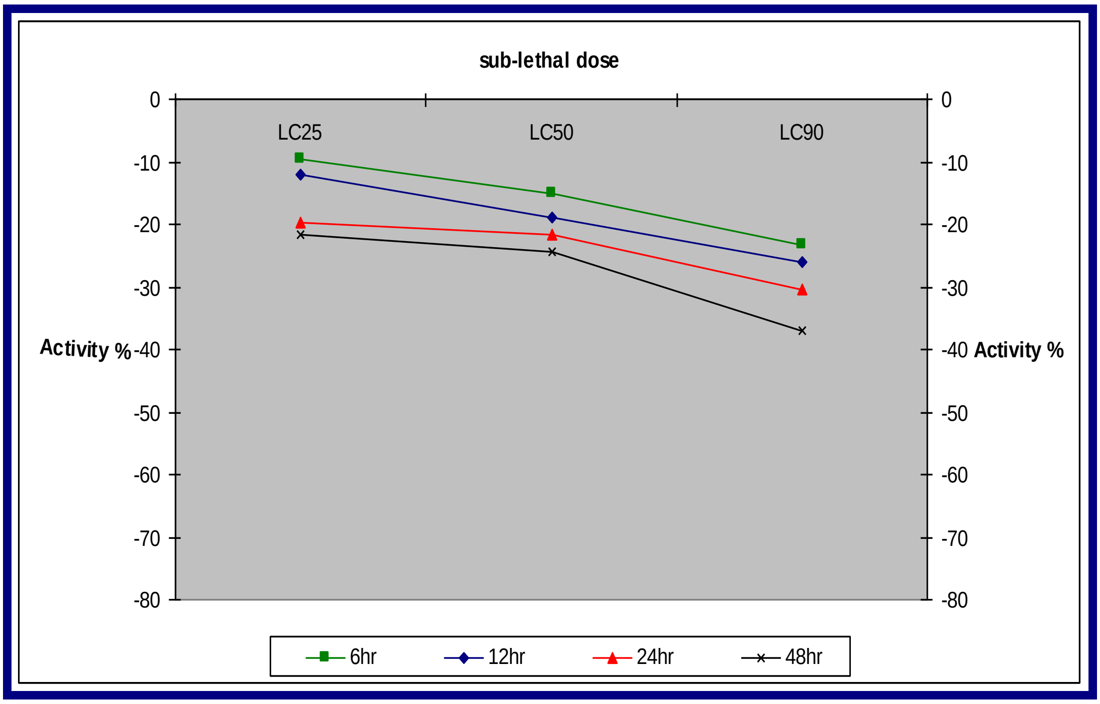


Fig. (33): β-esterase activity of 2nd larval instar treated with Sub-lethal doses of Match at different time intervals.

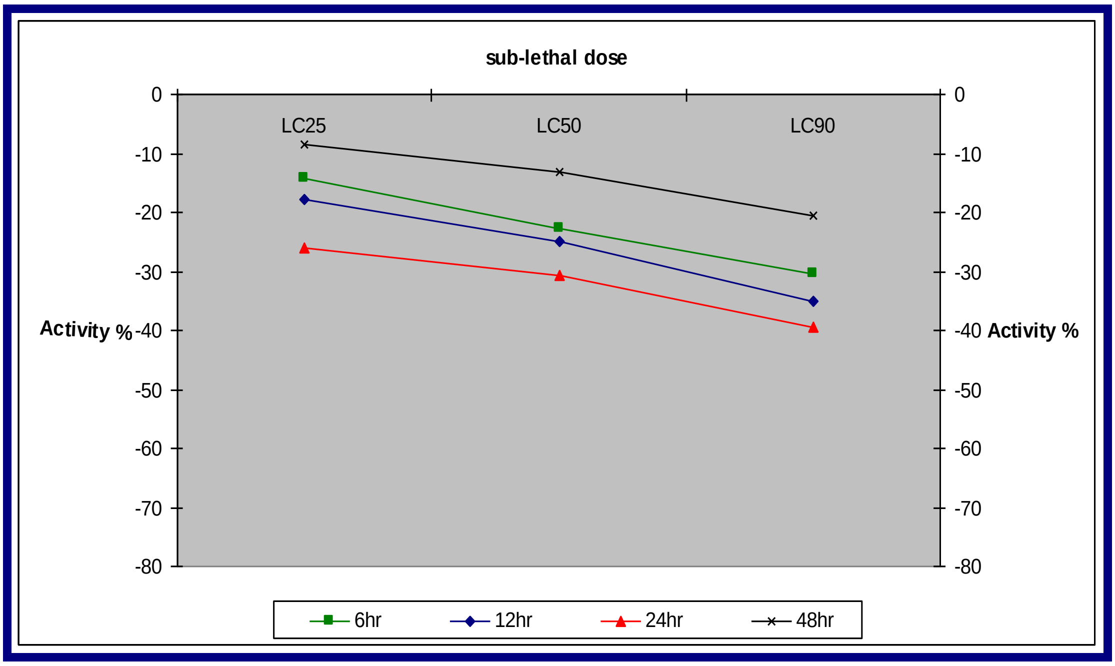


Fig. (34): β-esterase activity of 4th larval instar treated with Sub-lethal doses of Match at different time intervals.

4.4.5. Effect of Mimic on acid phosphatase :-

Results recorded in **Table (21)** and **Fig. (35)** showed that when 2nd larval instar was treated with sub-lethal concentrations the enzymatic activities were inhibited with the increase in the time post-treatment and also with the increase in dose.

Data obtained in **Table (21)** and **Fig. (36)** declared that treated 4th larval instar with sub-lethal concentrations showed a significant decreasing in activity with increasing of time post- treatment and increasing of dose.

4.4.6. Effect of Mimic on non-specific esterases(α, β- esterases):-

Results obtained and tabulated in **Tables (22)&(23)** and illustrated in **Figures (37)&(39)** declared that treated 2nd larval instar showed an decreasing in enzymatic activities as the dose increase and the time post-treatment increase.

Treated 4th larval instar with sub-lethal doses showed an reduction activities of non-specific esterases as the time post-treatment increase and also with the increasing of the dose, as showed in **Tables (22)&(23)** and illustrated in **Figures (38)&(40)**.

Table (21):- Acid phosphatase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Mimic at different time intervals.

Larval stage	Dose (ppm)	Acid phosphatase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC$_{25}$ (1.1)	6	2.0 ±0.01	1.92 ±0.6**	-4
		12	9.445 ±0.2	7.83 ±0.44**	-17.099
		24	11.86 ±0.24	9.73 ±0.31**	-17.96
		48	13.47 ±0.41	11.64 ±0.65**	-13.9
	LC$_{50}$ (1.5)	6	2.0 ±0.01	1.43 ±0.2**	-28.5
		12	9.445 ±0.2	5.98 ±0.71**	-36.69
		24	11.86 ±0.24	6.89 ±0.21**	-41.9
		48	13.47 ±0.41	7.94 ±0.62**	-41.05
	LC$_{90}$ (2.5)	6	2.0 ±0.01	1.0 ±0.04**	-50
		12	9.445 ±0.2	4.75 ±0.25**	-49.71
		24	11.86 ±0.24	5.18 ±0.31**	-56.32
		48	13.47 ±0.41	6.34 ±0.45**	-52.93
4th larval instar	LC$_{25}$ (6.1)	6	5.979 ±0.05	5.84 ±0.7**	-2.32
		12	22.89 ±0.38	20.74 ±0.47**	-9.4
		24	23.73 ±0.42	19.48 ±0.64**	-17.91
		48	26.94 ±0.53	21.15 ±0.22**	-21.49
	LC$_{50}$ (8.4)	6	5.979 ±0.05	5.11 ±0.28**	-14.53
		12	22.89 ±0.38	17.56 ±0.69**	-23.28
		24	23.73 ±0.42	16.77 ±0.84**	-29.33
		48	26.94 ±0.53	17.96 ±0.62**	-33.33
	LC$_{90}$ (15)	6	5.979 ±0.05	4.23 ±0.14**	-29.25
		12	22.89 ±0.38	15.92 ±0.43**	-30.45
		24	23.73 ±0.42	14.84 ±0.51**	-37.46
		48	26.94 ±0.53	15.83 ±0.42**	-41.24

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

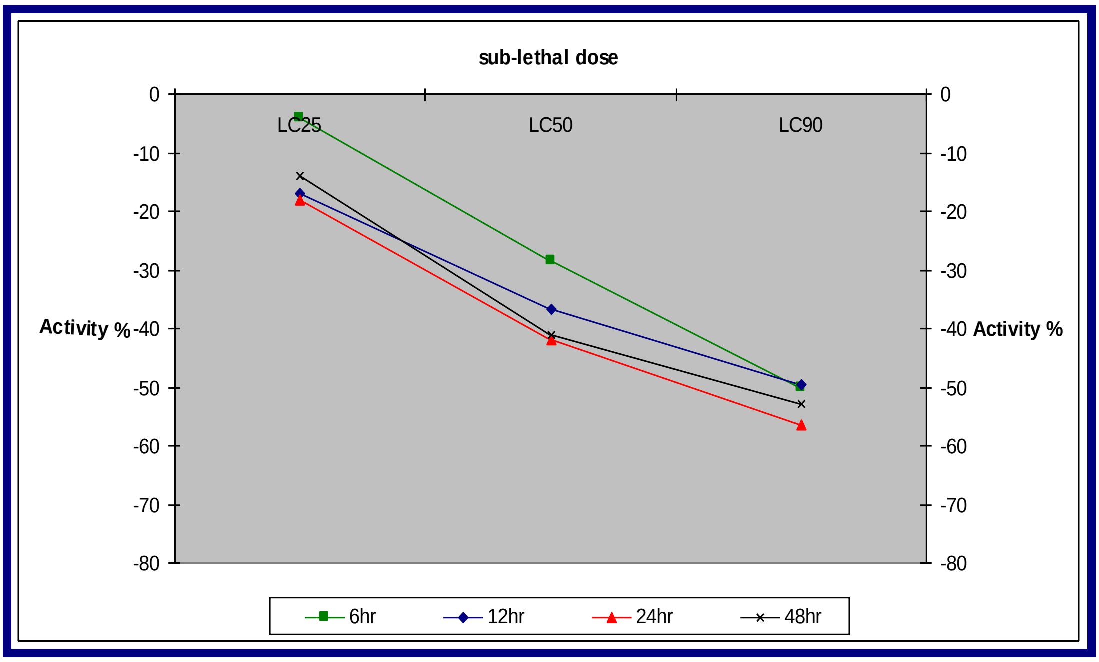


Fig. (35): Acid phosphatase activity of 2nd larval instar treated with Sub-lethal doses of Mimic at different time intervals.

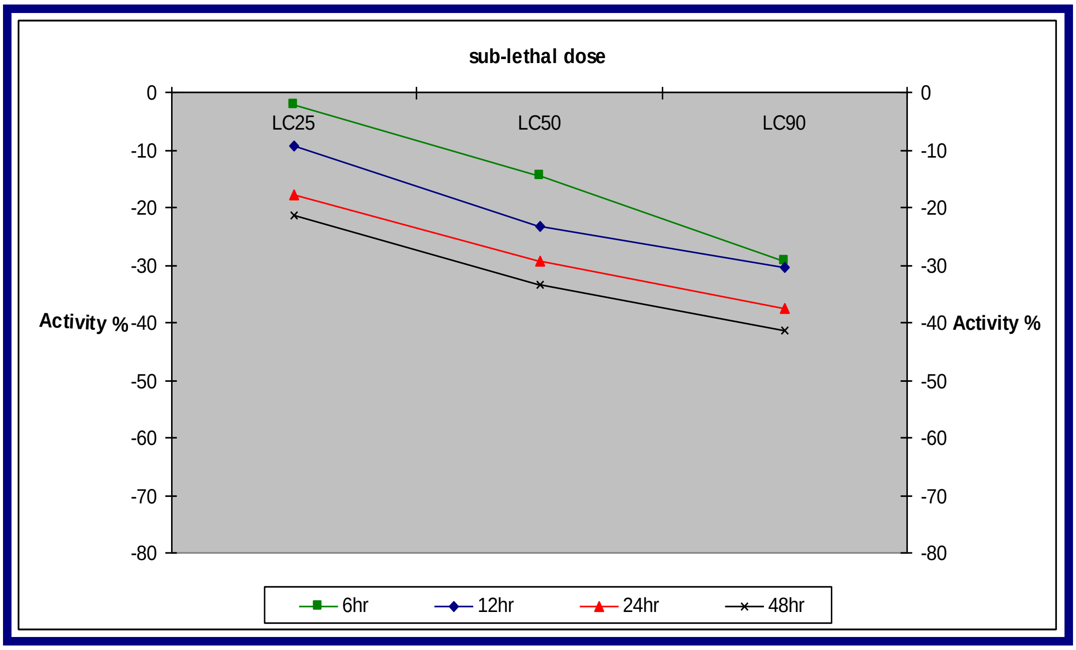


Fig. (36): Acid phosphatase activity of 4th larval instar treated with Sub-lethal doses of Mimic at different time intervals.

Table (22): α-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Mimic at different time intervals.

Larval stage	Dose (ppm)	α-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC$_{25}$ (1.1)	6	464.835 ±2.64	406.964 ±4.7	-12.45
		12	474.665 ±1.9	401.673 ±4.1*	-15.38
		24	553.33 ±2.4	447.952 ±3.6*	-19.04
		48	673.335 ±3.4	522.825 ±2.1**	-20.35
	LC$_{50}$ (1.5)	6	464.835 ±2.64	387.493 ±2.4*	-16.64
		12	474.665 ±1.9	368.762 ±3.7**	-20.31
		24	553.33 ±2.4	434.886 ±1.6**	-21.41
		48	673.335 ±3.4	509.746 ±4.3**	-24.3
	LC$_{90}$ (2.5)	6	464.835 ±2.64	341.638 ±4.1**	-26.5
		12	474.665 ±1.9	335.593 ±2.6**	-29.3
		24	553.33 ±2.4	400.937 ±3.5**	-27.54
		48	673.335 ±3.4	453.549 ±5.2**	-32.64
4th larval instar	LC$_{25}$ (6.1)	6	749.57 ±3.6	697.48 ±4.2*	-6.95
		12	786.43 ±1.8	653.89 ±5.1*	-16.85
		24	899.67 ±2.8	756.96 ±3.2**	-15.86
		48	976.83 ±2.4	800.67 ±2.2**	-18.03
	LC$_{50}$ (8.4)	6	749.57 ±3.6	652.52 ±3.4*	-12.95
		12	786.43 ±1.8	629.98 ±5.6*	-19.89
		24	899.67 ±2.8	701.83 ±1.2**	-21.99
		48	976.83 ±2.4	735.64 ±4.1**	-24.69
	LC$_{90}$ (15)	6	749.57 ±3.6	621.75 ±1.6**	-17.05
		12	786.43 ±1.8	592.93 ±1.2**	-24.6
		24	899.67 ±2.8	647.67 ±4.1**	-28.01
		48	976.83 ±2.4	683.38 ±1.1**	-30.04

*** Significant at P = 0.05** **** High significant at P = 0.01**

***** Very high significant at P = 0.001**

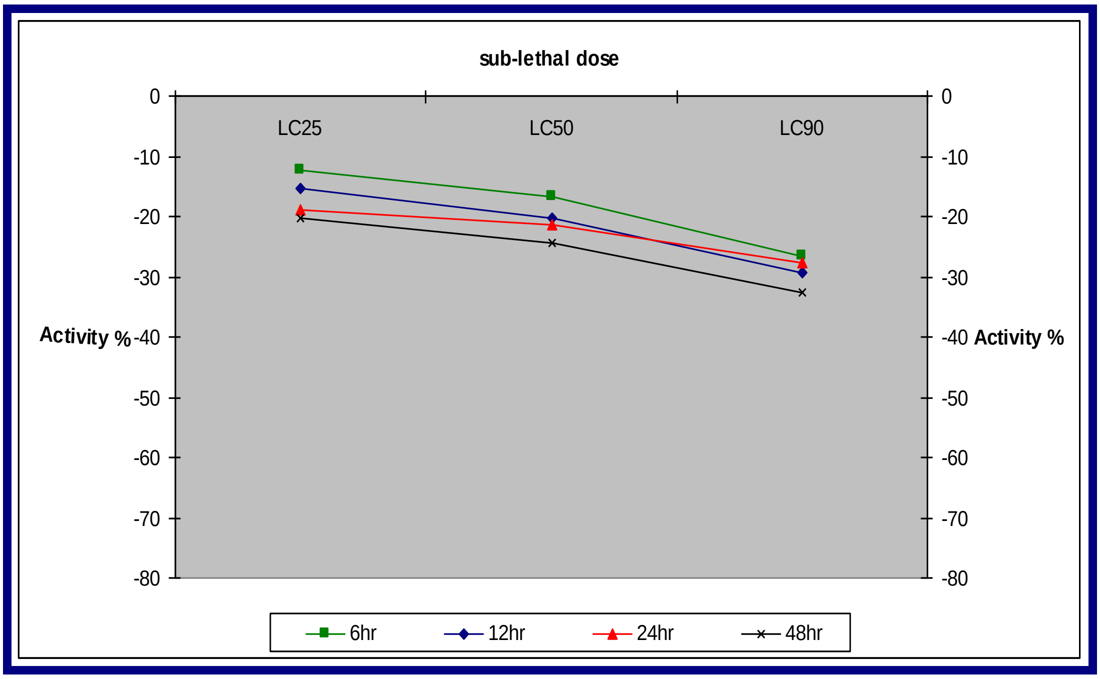


Fig. (37): α-esterase activity of 2nd larval instar treated with Sub-lethal doses of Mimic at different time intervals.

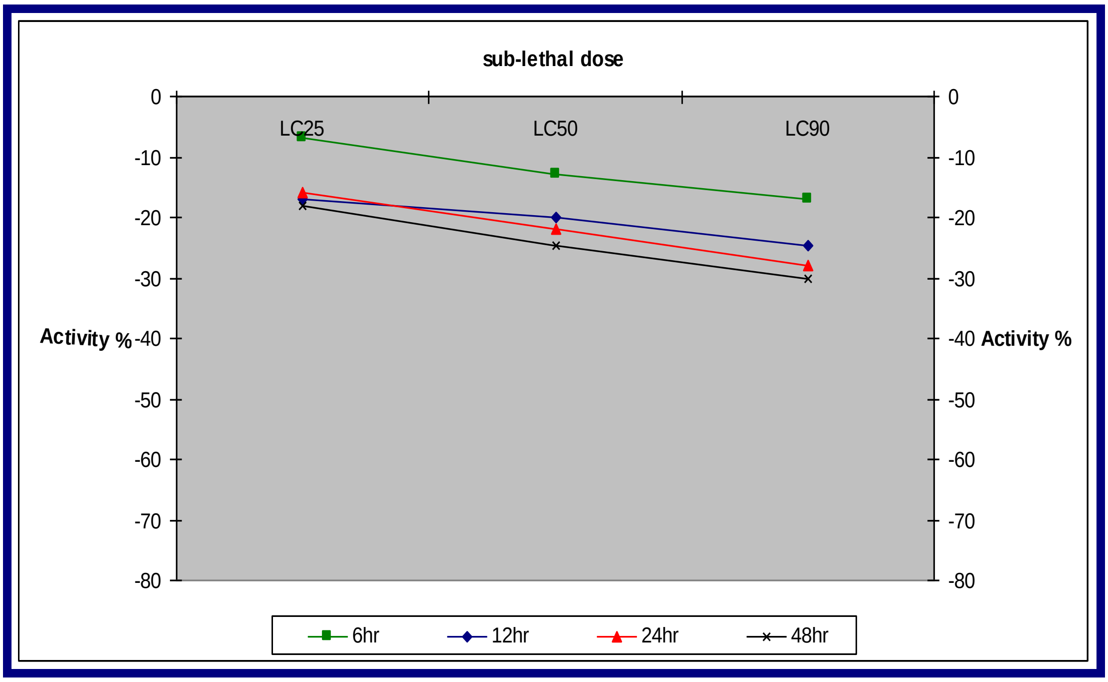


Fig. (38): α-esterase activity of 4th larval instar treated with Sub-lethal doses of Mimic at different time intervals.

Table (23): β-Esterase activity of 2nd and 4th larval instars treated with sub-lethal concentrations of Mimic at different time intervals.

Larval stage	Dose (ppm)	β-Esterase activity (µg phenol released/b.wt./min) Mean ±SE			Activity (%)
		Hours post- treatment	Control	Treated	
2nd larval instar	LC_{25} (1.1)	6	619.165 ±5.2	586.2 ±11.7*	-5.32
		12	842.665 ±4.4	773.16 ±9.9*	-8.25
		24	956.5 ±7.6	805.33 ±14.3**	-15.8
		48	1148.33 ±9.3	930.29 ±10.2**	-18.99
	LC_{50} (1.5)	6	619.165 ±5.2	545.4 ±8.5**	-11.9
		12	842.665 ±4.4	698.73 ±9.7**	-17.1
		24	956.5 ±7.6	764.43 ±12.4**	-20.0
		48	1148.33 ±9.3	903.5 ±10.5*	-21.32
	LC_{90} (2.5)	6	619.165 ±5.2	512.6 ±8.7**	-17.21
		12	842.665 ±4.4	664.33 ±11.3**	-21.16
		24	956.5 ±7.6	731.41 ±9.9**	-23.53
		48	1148.33 ±9.3	800.51 ±15.4**	-30.29
4th larval instar	LC_{25} (6.1)	6	1267.33 ±22.3	1197.83 ±24.04*	-5.5
		12	1734.28 ±32.1	1602.33 ±29.5*	-7.6
		24	1913.42 ±17.4	1694.63 ±10.3**	-11.43
		48	1125.48 ±24.3	1100 ±32.1	-2.3
	LC_{50} (8.4)	6	1267.33 ±22.3	1150.84 ±33.6*	-9.2
		12	1734.28 ±32.1	1504.33 ±26.8*	-13.26
		24	1913.42 ±17.4	1564.8 ±19.02**	-18.22
		48	1125.48 ±24.3	1068.5 ±25.7	-5.06
	LC_{90} (15)	6	1267.33 ±22.3	1054.32 ±30.2**	-16.81
		12	1734.28 ±32.1	1380.9 ±28.5**	-22.7
		24	1913.42 ±17.4	1409.2 ±20.4**	-26.35
		48	1125.48 ±24.3	1013.6 ±27.3*	-9.9

* Significant at P = 0.05 ** High significant at P = 0.01

*** Very high significant at P = 0.001

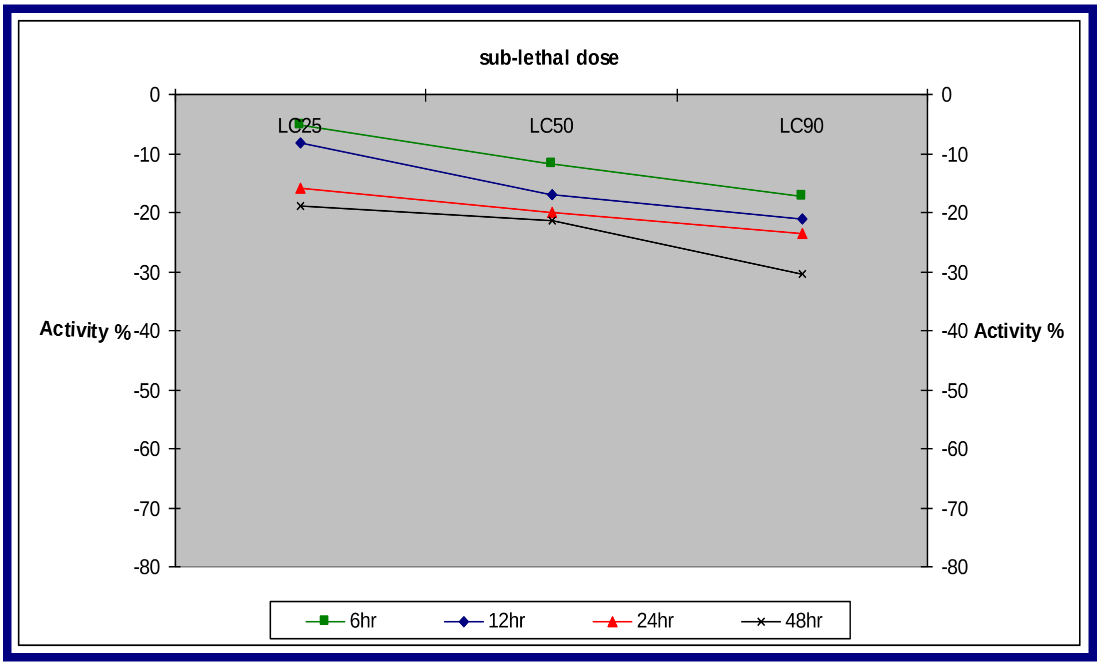


Fig. (39): β-esterase activity of 2nd larval instar treated with Sub-lethal doses of Mimic at different time intervals.

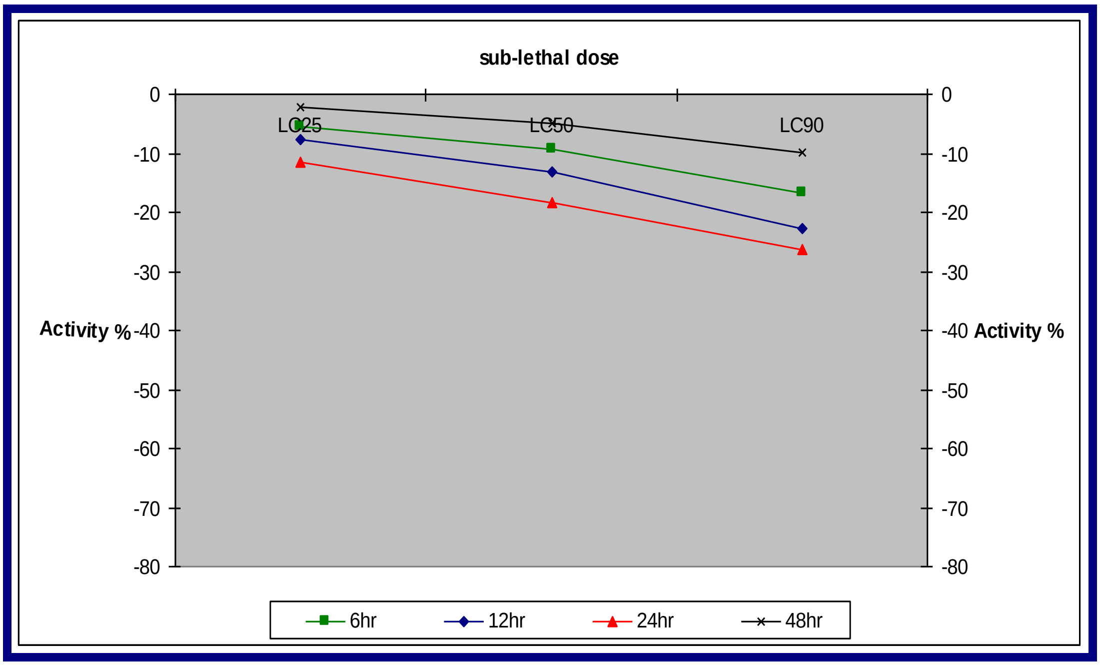


Fig. (40): β-esterase activity of 4th larval instar treated with Sub-lethal doses of Mimic at different time intervals.

4.5 -Molecular biological studies:-

PCR-RFLP technique was used to detect the various mutations in the nuclear vira-like chitinase gene of *Spodoptera littoralis* 2nd larval instar as a result of treatment with the different sub lethal concentrations (LC_{25}, LC_{50} and LC_{90}) at different time intervals post treatment (6, 12, 24 and 48hrs) . The resulting restriction fragments, which have been separated by agarose gel electrophoresis, were obtained as photos to enable us to detect mutations.

DNA genome has a higher molecular weight, so it remained in the wheel of the agarose gel electrophoresis **Plate (6)**. On the other hand DNA ladder represented in the 1st lane was separated into bands with different lengths.

The PCR products of vira-like chitinase gene for the different treated groups and the normal were found in lanes 1 to 10 respectively **Plate (7)**. The bands of vira-like chitinase gene for the different groups have the same length 1218bp.

The restricted fragments resulting from the digestion of vira-like chitinase gene of the different groups at the different time intervals post treatment with the enzyme ***SpeI*** are represented in **Plate (8)** in lanes 2 to10 respectively. The enzyme cut the vira-like chitinase gene one time and resulted in two bands with lengths 137pb and 1081bp, these bands are equal in the different studied groups as showed in **Table (24)** and illustrated in **Fig.(41)**.

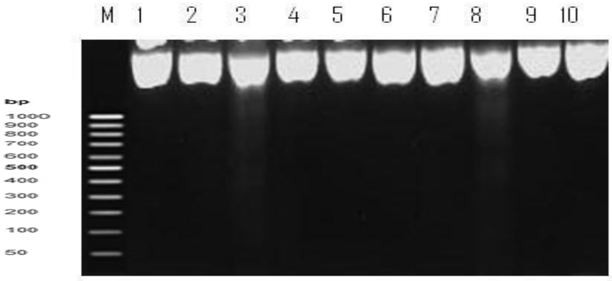


Plate (6): Shows the DNA ladder in the 1st lane while the other lanes contain the DNA extract of the total DNA genome for the studied groups all at the same length.

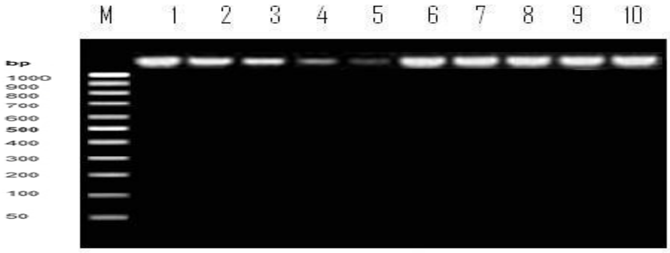


Plate (7): Shows the DNA ladder in the 1st lane while the other lanes contain the PCR products of the vira-like chitinase gene for the studied groups all at the same length.

M : Marker
2 : LC_{25} of Cascade
3: LC_{50} of Cascade
4: LC_{90} of Cascade

1 : Normal
5: LC_{25} of Match
6: LC_{50} of Match
7: LC_{90} of Match

8: LC_{25} of Mimic
9: LC_{50} of Mimic
10: LC_{90} of Mimic

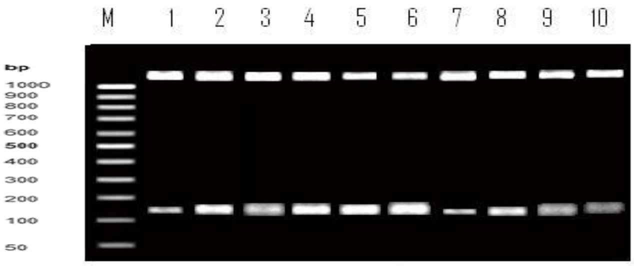


Plate (8): Represents that *SpeI* restricted the vira-like chitinase gene of all groups at the different time intervals post treatment with the same lengths 137pb and 1081pb.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	137	137	137	137	137	137	137	137	137	137
Band2	1081	1081	1081	1081	1081	1081	1081	1081	1081	1081

Table (24): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *SpeI* after 6,12,24 and 48hrs.

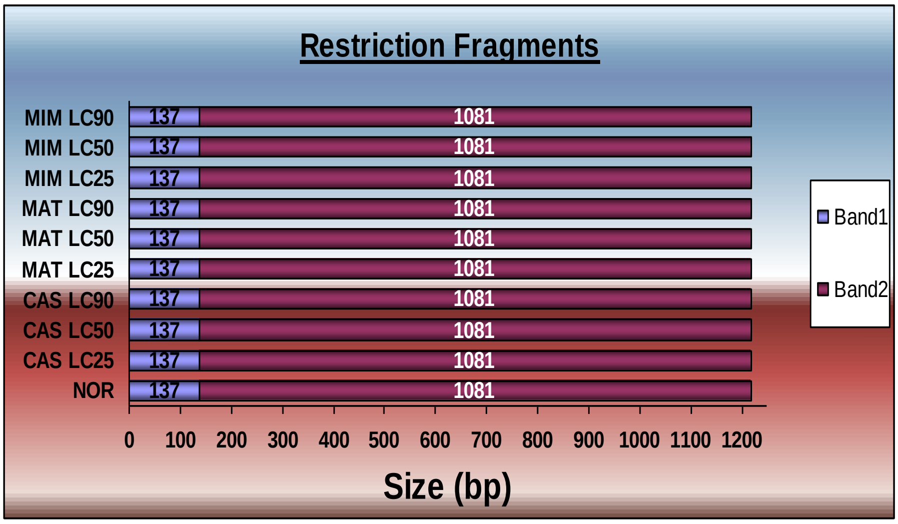


Fig. (41): Represents that *SpeI* restricted the vira-like chitinase gene of all groups at the different time intervals post treatment with the same lengths 137pb and 1081pb.

The enzyme ***EcoRI*** cut the vira-like chitinase gene of the treated larvae with sublethal doses LC_{50} and LC_{90} of Cascade as well as sublethal dose LC_{90} of Match after 6 hrs. post treatment **Plate (9)** lanes (3-4-7). Resulted in three bands with lengths 129pb, 334pb and 755pb, remaining different treated groups have the similar restricted fragments as normal with lengths 334pb and 884pb as showed in **Table (25)** and illustrated in **Fig.(42)**.

Plate (10) showed that the enzyme ***EcoR*I** cut the vira-like chitinase gene of the treated larvae with sub lethal doses LC_{50} and LC_{90} of Cascade, Match and Mimic after 12 hrs, lanes (3-4-6-7-9-10). Resulted in three bands with lengths 129pb, 334pb and 755pb, remaining different treated groups have the similar restricted fragments as normal with lengths 334pb and 884pb as showed in **Table (26)** and illustrated in **Fig.(43)**.

Plate (11) shows the PCR-RFLP products of the studied gene of the different studied groups after 24&48hrs. post treatment, which restricted with the enzyme ***EcoRI*** lanes (2-3-4-5-6-7-8-9-10)**.** Data tabulated in **Table (27)** and illustrated in **Fig.(44)** showed that it cut the vira-like chitinase gene two times resulting in three restricted fragments. These restricted fragments are equal in the different groups with lengths 129pb, 334pb and 755pb, lanes 2 to 10.

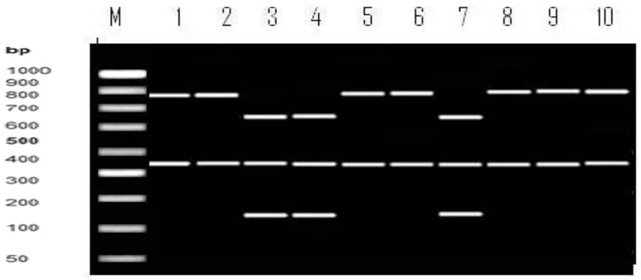


Plate (9): Shows the digestion of enzyme *EcoRI* to the vira-like chitinase gene of all groups at 6hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	334	334	129	129	334	334	129	334	334	334
Band2	884	884	334	334	884	884	334	884	884	884
Band3			755	755			755			

Table (25): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *EcoRI* after 6hrs.

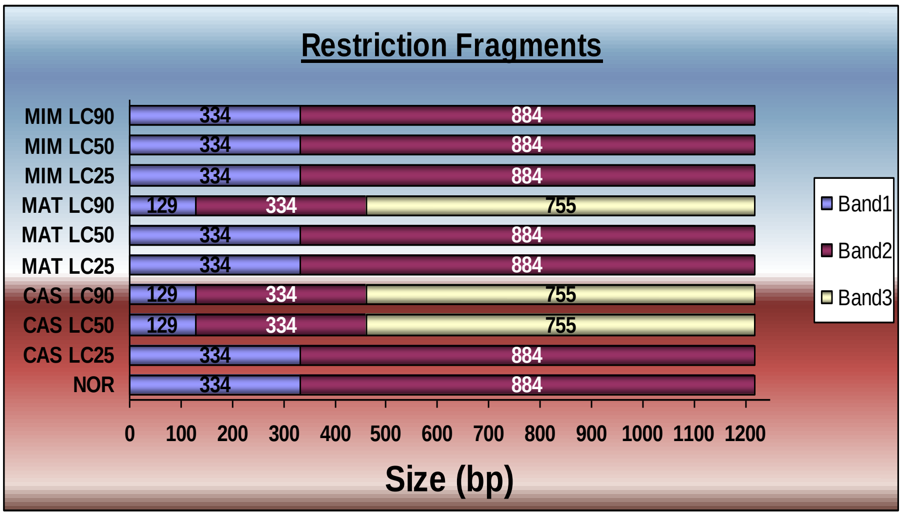


Fig. (42): Shows the digestion of enzyme *EcoRI* to the vira-like chitinase gene of all groups at 6hrs. post treatment.

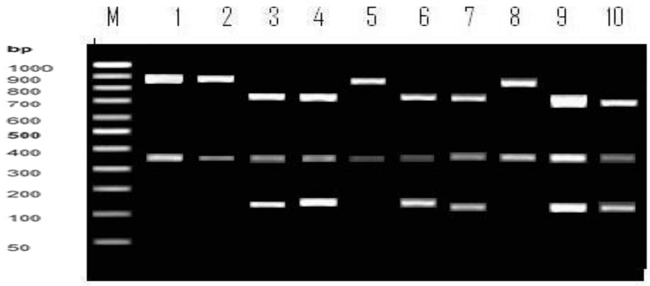


Plate (10): Shows the digestion of enzyme *EcoRI* to the vira-like chitinase gene of all groups at 12hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	334	334	129	129	334	129	129	334	129	129
Band2	884	884	334	334	884	334	334	884	334	334
Band3			755	755		755	755		755	755

Table (26): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *EcoRI* after 12hrs.

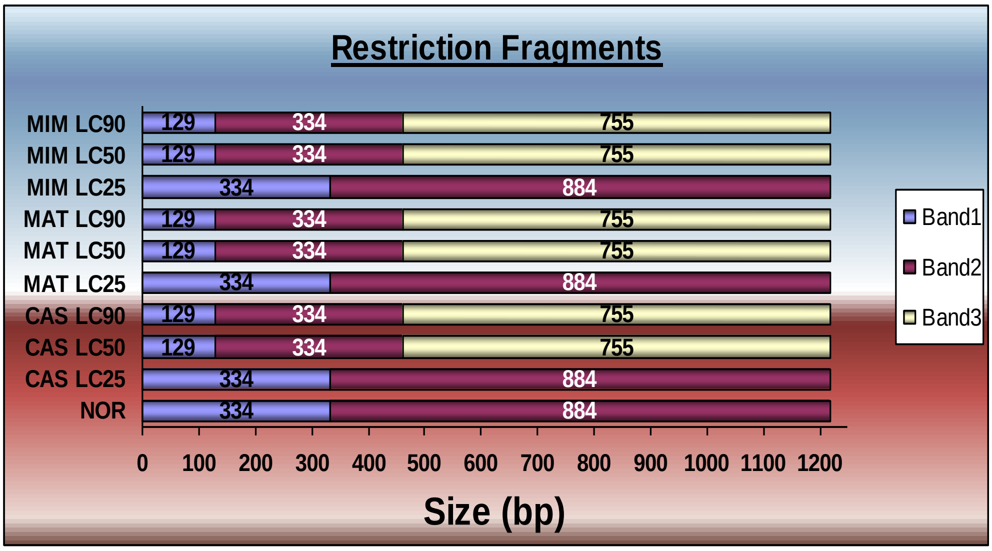


Fig. (43): Shows the digestion of enzyme *EcoRI* to the vira- like chitinase gene of all groups at 12hrs. post treatment.

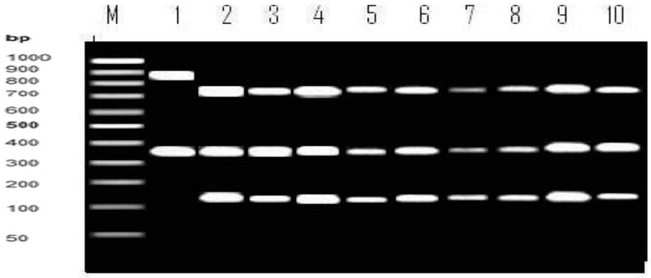


Plate (11): Shows the digestion of enzyme *EcoRI* to the vira-like chitinase gene of all groups at 24 &48 hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	334	129	129	129	129	129	129	129	129	129
Band2	884	334	334	334	334	334	334	334	334	334
Band3		755	755	755	755	755	755	755	755	755

Table (27): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *EcoRI* after 24& 48hrs.

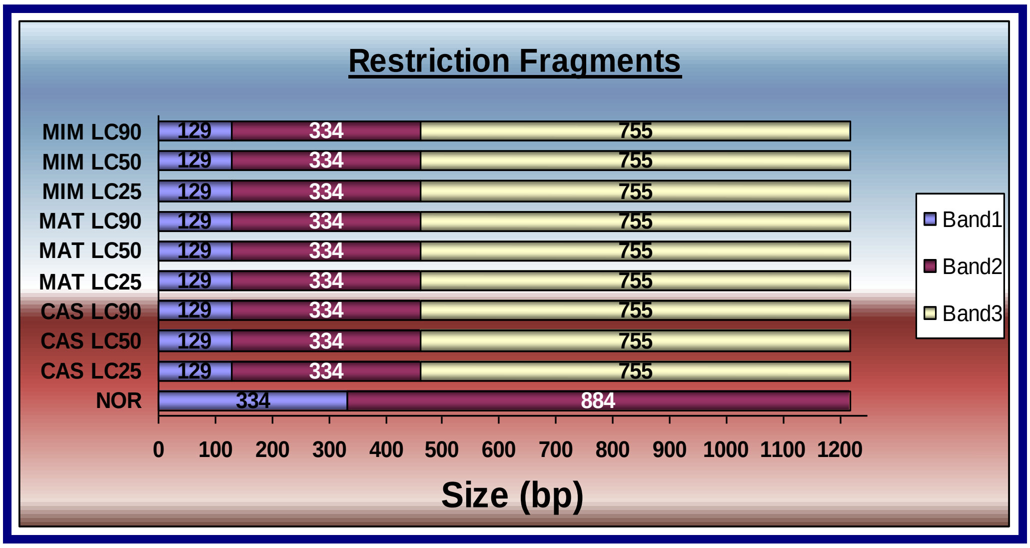


Fig. (44): Shows the digestion of enzyme *EcoRI* to the vira- like chitinase gene of all groups at 24&48 hrs. post treatment.

The enzyme ***BbsI*** cut the vira-like chitinase gene of the treated larvae with sub lethal doses LC_{50} and LC_{90} of Cascade as well as sub lethal dose LC_{90} of Match and Mimic after 6 hrs. post treatment **Plate (12)** lanes (3-4-7-10). Producing three bands these bands with lengths 60pb, 134pb and 1024pb, remaining groups as well as normal has the same lengths 60pb and 1158pb as showed in **Table (28)** and illustrated in **Fig.(45)**.

Plate (13) showed the restricted fragments, which resulted from the digestion of vira-like chitinase gene of the treated larvae with sub lethal doses after 12hrs. post treatment with the enzyme ***BbsI*.** Data tabulated in **Table (29)** and illustrated in **Fig.(46)** showed that, three distinct bands were presented in lanes (3-4) at lengths 60pb,134pb and 1024pb as a result of the restriction of vira-like chitinase gene of the treated larvae with the sub lethal doses LC_{50} and LC_{90} of Cascade. Vira-like chitinase gene of the Treated larvae with the sub lethal doses LC_{50} and LC_{90} of Match**,** were restricted into three distinct bands in lanes (6-7) at lengths 60pb, 517pb and 641pb. Vira-like chitinase gene of the Larvae treated with the sub lethal doses LC_{50} and LC_{90} of Mimic were cut into three distinct bands were presented in lanes (9-10) at lengths 60pb,324pb and 834pb.

Plate (14) showed the digestion of Vira-like chitinase gene after treatment of larvae with the different sub lethal doses of the different IGRs for 24&48hrs. with the enzyme ***BbsI***. Data tabulated in **Table (30)** and illustrated in **Fig.(47)** showed that, three distinct bands were presented in lane 2 at lengths 60pb, 134pb and 1024pb as a result of the restriction of vira-like chitinase gene of the treated larvae with the sub lethal dose LC_{25} of Cascade. In lanes (3-4) there were four bands at lengths 60pb, 134pb, 488pb and 536 pb represents the result of restriction of vira-like chitinase gene of the treated larvae with the sub lethal doses LC_{50} and LC_{90} of Cascade. In lane 5 there were three bands at lengths 60pb, 517pb and 641pb as a result of the restriction of vira-like chitinase gene of the treated larvae with the sub lethal dose LC_{25} of Match. The enzyme ***BbsI*** cut the vira-like chitinase gene of the treated larvae with sub lethal doses LC_{50} and LC_{90} of Match after 24&48hrs. post treatment represented in lanes (6-7) three times resulting in four bands with lengths 60pb, 200pb, 441pb and 517pb. The last three sub lethal doses of the last IGR Mimic have the same restricted fragments when cut with the same enzyme at the lengths 60pb,324pb and 834pb and represented in lanes (8-9-10).

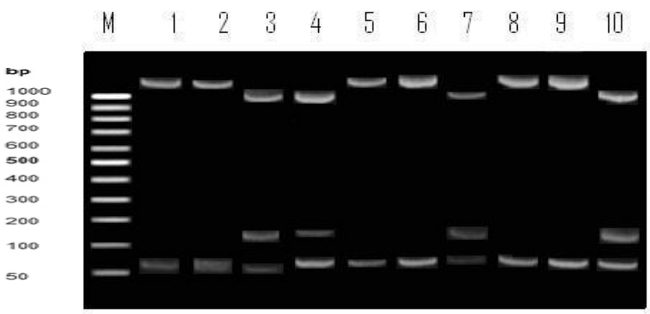


Plate (12): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 6hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	60	60	60	60	60	60	60	60	60	60
Band2	1158	1158	134	134	1158	1158	134	1158	1158	134
Band3			1024	1024			1024			1024

Table (28): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *BbsI* after 6hrs.

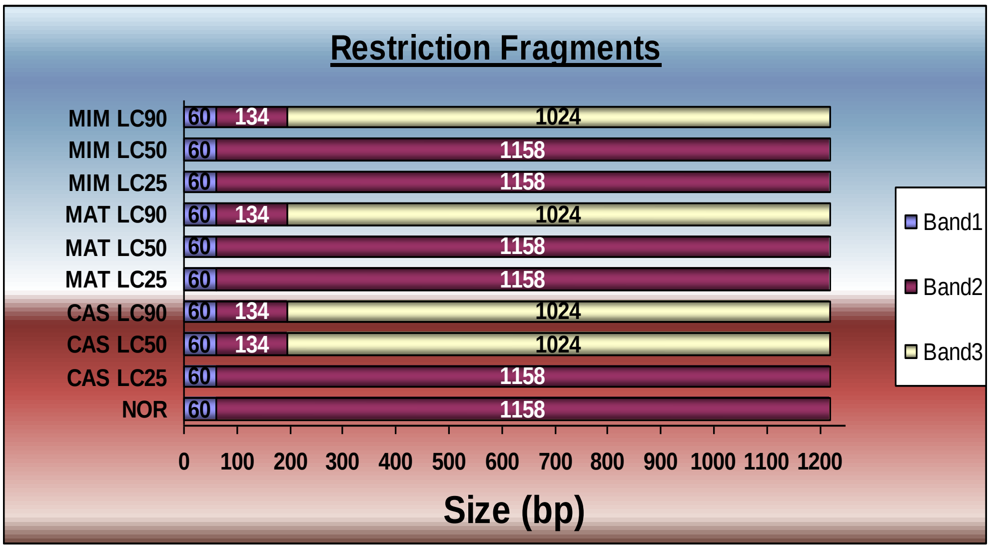


Fig. (45): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 6hrs. post treatment.

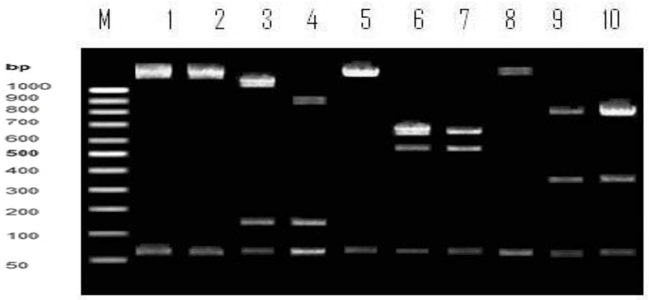


Plate (13): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 12hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	60	60	60	60	60	60	60	60	60	60
Band2	1158	1158	134	134	1158	517	517	1158	324	324
Band3			1024	1024		641	641		834	834

Table (29): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *BbsI* after 12hrs.

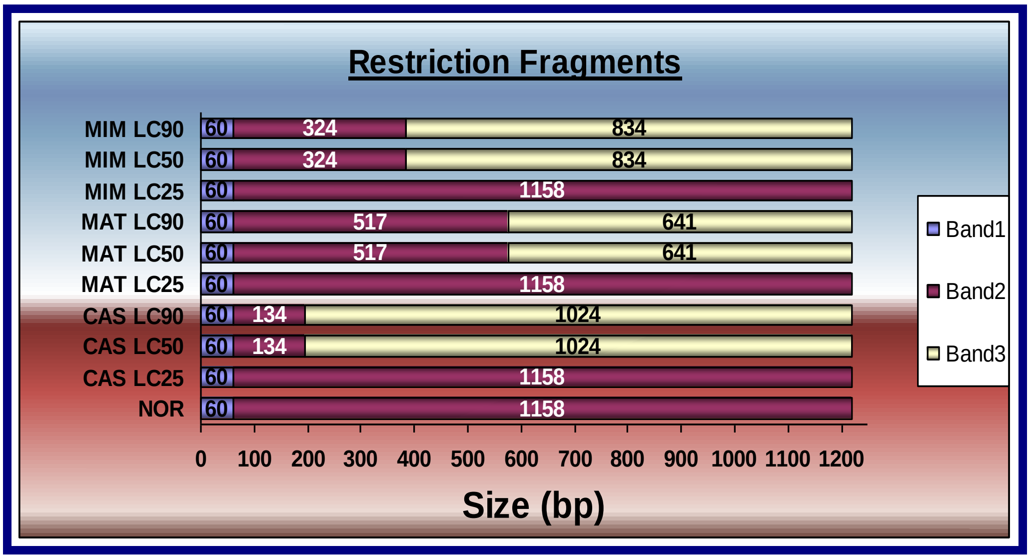


Fig. (46): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 12hrs. post treatment.

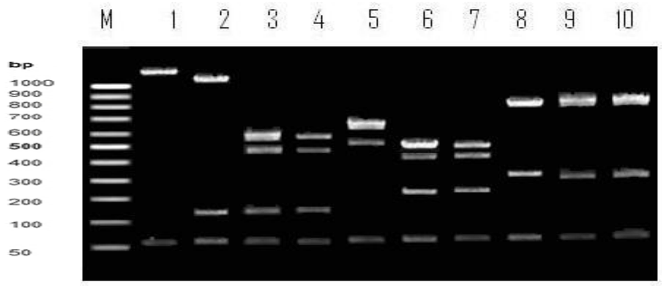


Plate (14): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 24&48 hrs. post treatment.

	NOR	CAS LC25	CAS LC50	CAS LC90	MAT LC25	MAT LC50	MAT LC90	MIM LC25	MIM LC50	MIM LC90
Band1	60	60	60	60	60	60	60	60	60	60
Band2	1158	134	134	134	517	200	200	324	324	324
Band3		1024	488	488	641	441	441	834	834	834
Band4			536	536		517	517			

Table (30): Shows the lengths of vira-like chitinase gene fragments, which restricted with the endonuclease *BbsI* after 24& 48hrs.

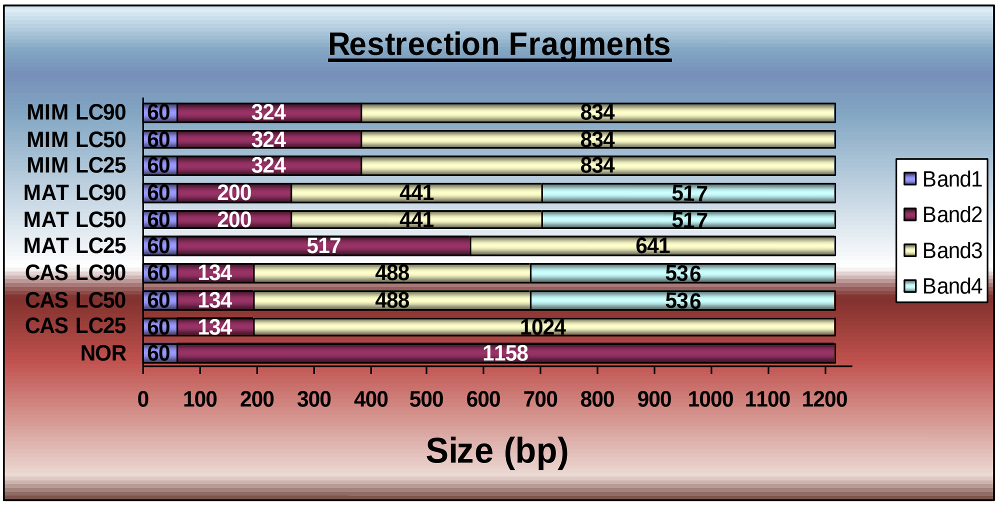


Fig. (47): Shows the digestion of enzyme *BbsI* to the vira-like chitinase gene of all groups at 24&48 hrs. post treatment.

4.5.1. Bioinformatics:-

In the present study the sequence of the *Spodoptera littoralis* vira-like chitinase gene, 1218pb, 398 encoded amino acids and 8 stop codons **Fig.(48)** and **Fig.(49),** respectively was used to determine the predicted mutations as a result of the treatment of 2nd larval instar with the different sub lethal doses of the different IGRs. All the details of the normal *Spodoptera littoralis* vira-like chitinase gene such as, percentage of bases, codons and amino acids …etc are represented in **Table (31)** and **Table (32),** respectively.

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTA
TCCACGATCCTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAA
CTTCGGCCAATTGATGATGTTGAAACAAGCAAGACCAGATCTGAAGATCTTGCCTTCTGTTGGTGGATGGACC
CTGGCTGATCCGTTCTTCTTTTTCACTGACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCC
TTGAAACCTGGAAGTTCTTTGATGGTGTTGACATTGATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGA
TCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATGAAGGAACTCAGAGAGATGTTGAACGAGCTT
TCTGCCAAGACTGGTAAGAAGTACGAATTGACATCTGCTATCAGCTCTGGTTGGGACAAGATTCAGGTTGTGG
ACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAGGGAGCTTGGTCTAA
CGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTGACTTT
GGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGATCGTTGTTGGTGTTGCAATGTATGGAAGAG
GTTGGACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAAC
TTGGCAGGATGGTGTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTAT
GATAAGGTGGCTCAGGCACCGTATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGAT
CGACAATTGAGAAAGCTAAATATGTGAGGAACAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGA
CAATGGTGACATATTAAATGCCATGAATATGGGTCTCGGTAACAATGCGTGAAGGGAAACTAGTGCGAGTGTT
GTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATGTCTCAAATTTTTATTTACAAAAATTG
TTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (48): Original sequence of *S. littoralis* vira-like gene.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKILPSVGGWT
LADPFFFFTDEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNEL
SAKTGKKYELTSAISSGWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDF
GVKFLLAQGVSPKKIVVGVAMYGRGWTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHY
DKVAQAPYVFRKETGDLITYDDARSTIEKAKYVRNNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASV
VCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KKSL
```

Fig. (49): Encoded amino acides of *S. littoralis* vira-like gene.

(∗) refers to stop codon

Sequence information:-

Sequence length: 1218 bases
Total codons: 406
Stop codons (*): 8
Encoded amino acids: 398 amino acids
Isoelectric point (I.P.): 7.1
Average MW (AMW): 44738.1
Monoisotopic MW (MMW): 44710.4795

Percentage of the nitrogenous bases:

A: 342 (28.08 %)
G: 327 (26.85 %)
AT = 54.85%

T: 326 (26.77 %)
C: 223 (28.31 %)
CG = 45.16%

	A	T	G	C
AA	16 (3.94%)	9 (2.22%)	20 (4.93%)	9 (2.22%)
AT	2 (0.49%)	8 (1.97%)	9 (2.22%)	6 (1.48%)
AG	7 (1.72%)	5 (1.23%)	4 (0.99%)	3 (0.74%)
AC	4 (0.99%)	11 (2.71%)	1 (0.25%)	2 (0.49%)
TA	2 (0.49%)	7 (1.72%)	2 (0.49%)	9 (2.22%)
TT	6 (1.48%)	5 (1.23%)	10 (2.46%)	16(3.94%)
TG	4 (0.99%)	4 (0.99%)	12 (2.96%)	1 (0.25%)
TC	2 (0.49%)	7 (1.72%)	2 (0.49%)	1 (0.25%)
GA	13 (3.20%)	11 (2.71%)	10 (2.46%)	16 (3.94%)
GT	4 (0.99%)	12 (2.96%)	8 (1.97%)	5 (1.23%)
GG	7 (1.72%)	22 (5.42%)	1 (0.25%)	7 (1. 72%)
GC	5 (1.23%)	15 (3.69%)	4 (0.99%)	6 (1.48%)
CA	5 (1.23%)	0 (0.00%)	10(2.46%)	6 (1.48%)
CT	0 (0.00%)	6 (1.48%)	6 (1.48%)	4 (0.99%)
CG	1 (0.25%)	0 (0.00%)	0 (0.00%)	1 (0.25%)
CC	3 (0.74%)	7 (1.72%)	3 (0.74%)	2 (0.49%)

Table (31): percentage of the different codons

Ala (A)	30 (7.54 %)	**Gly (G)**	37 (9.30 %)
Cys (C)	5 (1.26 %)	**His (H)**	6 (1.51 %)
Asp (D)	27 (6.78 %)	**Ile (I)**	16 (4.02 %)
Glu (E)	23 (5.78 %)	**Lys (K)**	36 (9.05 %)
Phe (F)	21(5.28 %)	**Leu (L)**	32 (8.04 %)
Met (M)	9 (9.30 %)	**Ser (S)**	20 (5.03 %)
Asn (N)	18. (4.52 %)	**Thr (T)**	18 (4.52 %)
Pro (P)	15 (3.77 %)	**Val (V)**	29 (7.29 %)
Gln (Q)	15 (3.77 %)	**Trp (W)**	12 (3.02 %)
Arg (R)	13 (3.27 %)	**Tyr (Y)**	16 (4.02 %)

Table (32): percentage of the encoded amino acides.

The following section is concerned with the mutations of the exposed groups of larvae which treated with the different sub lethal doses (LC_{25}-LC_{50}-LC_{90}) of the used IGRs (Cascade-Match-Mimic**)** at the different times intervals(6,12,24,48hrs). The light has been focused on the changes that occur in the sequence of the gene, the expressed protein and other parameters.

The treated 2nd larval instar with LC_{50} and LC_{90} of Cascade and LC_{90} of Match showed two mutations after 6hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.194)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, **Fig.(50)**. Therefore according to these two mutations the encoded amino acids showed two mutations. The first one involved the substitution of the amino acid **I (No.65)** with **T**, the second involved the substitution of the amino acid **L (No.156)** with **F**, **Fig.(51)**.

Exposed 2nd larval instar to LC_{90} of Mimic showed one mutation after 6hrs post treatment. Involved the substitution of the nitrogenous base **T (No.194)** with **C, Fig.(52) .** Therefore according to this mutation the encoded amino acids showed one mutation. Involved the substitution of the amino acid **I (No.65)** with **T**, **Fig.(53)**.

Accordingly, the following parameters have been changed**,** total codones numbers which represented in **table (33)** and amino acid numbers which represented in **table (34).**

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGAT►CCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCA
CTGACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACAT
TGATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATG
AAGGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTG►CACATCTGCTATCAGCTCTGG
TTGGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAG
GGAGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTG
ACTTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGATCGTTGTTGGTGTTGCAATGTATGGAAGAGGTTG
GACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATGGT
GTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACCGT
ATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGGAA
CAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCGGT
AACAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATGTC
TCAAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (50): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Cascade and LC_{90} of Match after 6hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKI►TLPSVGGWTLADPFFF
FTDEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYEL►FTSAI
SSGWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKIVVGVAMYG
RGWTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKAKY
VRNNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KKSL
```

Fig. (51): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Cascade and LC_{90} of Match after 6hrs.

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGAT►CCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCA
CTGACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACAT
TGATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATG
AAGGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTGACATCTGCTATCAGCTCTGGTT
GGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAGGG
AGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTGAC
TTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGATCGTTGTTGGTGTTGCAATGTATGGAAGAGGTTGGA
CTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATGGTGT
GGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACCGTAT
GTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGGAACA
ATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCGGTAA
CAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATGTCTC
AAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (52): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{90} of Mimic after 6hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKI►TLPSVGGWTLADPFFF
FTDEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYELTSAISS
GWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKIVVGVAMYGRG
WTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKAKYVR
NNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KKSL
```

Fig. (53): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{90} of Mimic after 6hrs.

Table (33): change in total number of codons after 6hrs. of treatment.

codons	Number of codons									
	NOR.	CAS. LC_{25}	CAS. LC_{50}	CAS. LC_{90}	MAT. LC_{25}	MAT. LC_{50}	MAT. LC_{90}	MIM. LC_{25}	MIM. LC_{50}	MIM. LC_{90}
TTT	5	5	5	5	5	5	5	5	5	5
TTC	16	16	17	17	16	16	17	16	16	16
TTA	6	6	6	6	6	6	6	6	6	6
TTG	10	10	9	9	10	10	9	10	10	10
CTT	6	6	6	6	6	6	6	6	6	6
CTC	4	4	4	4	4	4	4	4	4	4
CTA	0	0	0	0	0	0	0	0	0	0
CTG	6	6	6	6	6	6	6	6	6	6
ATT	8	8	8	8	8	8	8	8	8	8
ATC	6	6	5	5	6	6	5	6	6	5
ATA	2	2	2	2	2	2	2	2	2	2
ATG	9	9	9	9	9	9	9	9	9	9
GTT	12	12	12	12	12	12	12	12	12	12
GTC	5	5	5	5	5	5	5	5	5	5
GTA	4	4	4	4	4	4	4	4	4	4
GTG	8	8	8	8	8	8	8	8	8	8
TCT	7	7	7	7	7	7	7	7	7	7
TCC	1	1	1	1	1	1	1	1	1	1
TCA	2	2	2	2	2	2	2	2	2	2
TCG	2	2	2	2	2	2	2	2	2	2
CCT	7	7	7	7	7	7	7	7	7	7
CCC	2	2	2	2	2	2	2	2	2	2
CCA	3	3	3	3	3	3	3	3	3	3
CCG	3	3	3	3	3	3	3	3	3	3
ACT	11	11	11	11	11	11	11	11	11	11
ACC	2	2	3	3	2	2	3	2	2	3
ACA	4	4	4	4	4	4	4	4	4	4
ACG	1	1	1	1	1	1	1	1	1	1
GCT	15	15	15	15	15	15	15	15	15	15
GCC	6	6	6	6	6	6	6	6	6	6
GCA	5	5	5	5	5	5	5	5	5	5
GCG	4	4	4	4	4	4	4	4	4	4

Continued table (33)

codons	Number of codons									
	NOR.	CAS. LC$_{25}$	CAS. LC$_{50}$	CAS. LC$_{90}$	MAT. LC$_{25}$	MAT. LC$_{50}$	MAT. LC$_{90}$	MIM. LC$_{25}$	MIM. LC$_{50}$	MIM. LC$_{90}$
TAT	7	7	7	7	7	7	7	7	7	7
TAC	9	9	9	9	9	9	9	9	9	9
TAA	2	2	2	2	2	2	2	2	2	2
TAG	2	2	2	2	2	2	2	2	2	2
CAT	0	0	0	0	0	0	0	0	0	0
CAC	6	6	6	6	6	6	6	6	6	6
CAA	5	5	5	5	5	5	5	5	5	5
CAG	10	10	10	10	10	10	10	10	10	10
AAT	9	9	9	9	9	9	9	9	9	9
AAC	9	9	9	9	9	9	9	9	9	9
AAA	16	16	16	16	16	16	16	16	16	16
AAG	20	20	20	20	20	20	20	20	20	20
GAT	11	11	11	11	11	11	11	11	11	11
GAC	16	16	16	16	16	16	16	16	16	16
GAA	13	13	13	13	13	13	13	13	13	13
GAG	10	10	10	10	10	10	10	10	10	10
TGT	4	4	4	4	4	4	4	4	4	4
TGC	1	1	1	1	1	1	1	1	1	1
TGA	4	4	4	4	4	4	4	4	4	4
TGG	12	12	12	12	12	12	12	12	12	12
CGT	0	0	0	0	0	0	0	0	0	0
CGC	1	1	1	1	1	1	1	1	1	1
CGA	1	1	1	1	1	1	1	1	1	1
CGG	0	0	0	0	0	0	0	0	0	0
ACT	5	5	5	5	5	5	5	5	5	5
AGC	3	3	3	3	3	3	3	3	3	3
AGA	7	7	7	7	7	7	7	7	7	7
AGG	4	4	4	4	4	4	4	4	4	4
GGT	22	22	22	22	22	22	22	22	22	22
GGC	7	7	7	7	7	7	7	7	7	7
GGA	7	7	7	7	7	7	7	7	7	7
GGG	1	1	1	1	1	1	1	1	1	1
TOTAL CODON	406	406	406	406	406	406	406	406	406	406

The treated 2nd larval instar with LC_{50} and LC_{90} of Cascade showed two mutations after 12hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.194)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, **Fig.(50)**. Therefore according to these two mutations the encoded amino acids showed two mutations. The first one involved the substitution of the amino acid **I (No.65)** with **T**, the second involved the substitution of the amino acid **L (No.156)** with **F**, **Fig.(51)**. Exposed 2nd larval instar to LC_{50} and LC_{90} of Match showed two mutations after 12hrs post treatment. The first one involved the substitution of the nitrogenous base **G(No.468)** with **C,** the second mutation involved the substitution of the nitrogenous base **T (No.701)** with **C Fig.(54) .** Therefore according to these two mutations the encoded amino acids showed two mutations. The first one involved the substitution of the amino acid **L (No.156)** with **F**, the second involved the substitution of the amino acid **I (No.234)** with **T**, **Fig.(55)**.

The treated 2nd larval instar with LC_{50} and LC_{90} of Mimic showed two mutations after 12hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.384)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, **Fig.(56)**. Therefore according to these two mutations the encoded amino acids showed one mutation, involved the substitution of the amino acid **L (No.156)** with **F**, **Fig.(57)**.

Accordingly, the following parameters have been changed**,** total codones numbers which represented in **table (35)** and amino acid numbers which represented in **table (36).**

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGATCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCACT
GACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACATTG
ATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATGAA
GGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTG►CACATCTGCTATCAGCTCTGGTT
GGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAGGG
AGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTGAC
TTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGAT►CCGTTGTTGGTGTTGCAATGTATGGAAGAGGTTG
GACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATGGT
GTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACCGT
ATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGGAA
CAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCGGT
AACAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATGTC
TCAAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (54): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Match after 12hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKILPSVGGWTLADPFFFFT
DEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYEL►FTSAISS
GWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKI►TVVGVAMYG
RGWTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKAKY
VRNNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KKSL
```

Fig. (55): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Match after 12hrs.

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGATCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCACT
GACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACATTG
ATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGAT►CGGTCACATCTACGTCCAGCTGATG
AAGGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTG►CACATCTGCTATCAGCTCTGG
TTGGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAG
GGAGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTG
ACTTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGATCGTTGTTGGTGTTGCAATGTATGGAAGAGGTTG
GACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATGGT
GTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACCGT
ATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGGAA
CAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCGGT
AACAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATGTC
TCAAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (56): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Mimic after 12hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKILPSVGGWTLADPFFFFT
DEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYEL►FTSAISS
GWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKIVVGVAMYGRG
WTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKAKYVR
NNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KKSL
```

Fig. (57): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Mimic after 12hrs.

Table (35): change in total number of codons after 12hrs. post treatment.

codons	Number of codons									
	NOR.	CAS. LC_{25}	CAS. LC_{50}	CAS. LC_{90}	MAT. LC_{25}	MAT. LC_{50}	MAT. LC_{90}	MIM. LC_{25}	MIM. LC_{50}	MIM. LC_{90}
TTT	5	5	5	5	5	5	5	5	5	5
TTC	16	16	17	17	16	17	17	16	17	17
TTA	6	6	6	6	6	6	6	6	6	6
TTG	10	10	9	9	10	9	9	10	9	9
CTT	6	6	6	6	6	6	6	6	6	6
CTC	4	4	4	4	4	4	4	4	4	4
CTA	0	0	0	0	0	0	0	0	0	0
CTG	6	6	6	6	6	6	6	6	6	6
ATT	8	8	8	8	8	8	8	8	8	8
ATC	6	6	5	5	6	5	5	6	6	6
ATA	2	2	2	2	2	2	2	2	2	2
ATG	9	9	9	9	9	9	9	9	9	9
GTT	12	12	12	12	12	12	12	12	12	12
GTC	5	5	5	5	5	5	5	5	5	5
GTA	4	4	4	4	4	4	4	4	4	4
GTG	8	8	8	8	8	8	8	8	8	8
TCT	7	7	7	7	7	7	7	7	7	7
TCC	1	1	1	1	1	1	1	1	1	1
TCA	2	2	2	2	2	2	2	2	2	2
TCG	2	2	2	2	2	2	2	2	2	2
CCT	7	7	7	7	7	7	7	7	7	7
CCC	2	2	2	2	2	2	2	2	2	2
CCA	3	3	3	3	3	3	3	3	3	3
CCG	3	3	3	3	3	3	3	3	3	3
ACT	11	11	11	11	11	11	11	11	11	11
ACC	2	2	3	3	2	3	3	2	2	2
ACA	4	4	4	4	4	4	4	4	4	4
ACG	1	1	1	1	1	1	1	1	1	1
GCT	15	15	15	15	15	15	15	15	15	15
GCC	6	6	6	6	6	6	6	6	6	6
GCA	5	5	5	5	5	5	5	5	5	5
GCG	4	4	4	4	4	4	4	4	4	4

Continued table (35)

Codons	Number of codons									
	NOR.	CAS. LC_{25}	CAS. LC_{50}	CAS. LC_{90}	MAT. LC_{25}	MAT. LC_{50}	MAT. LC_{90}	MIM. LC_{25}	MIM. LC_{50}	MIM. LC_{90}
TAT	7	7	7	7	7	7	7	7	7	7
TAC	9	9	9	9	9	9	9	9	9	9
TAA	2	2	2	2	2	2	2	2	2	2
TAG	2	2	2	2	2	2	2	2	2	2
CAT	0	0	0	0	0	0	0	0	0	0
CAC	6	6	6	6	6	6	6	6	6	6
CAA	5	5	5	5	5	5	5	5	5	5
CAG	10	10	10	10	10	10	10	10	10	10
AAT	9	9	9	9	9	9	9	9	9	9
AAC	9	9	9	9	9	9	9	9	9	9
AAA	16	16	16	16	16	16	16	16	16	16
AAG	20	20	20	20	20	20	20	20	20	20
GAT	11	11	11	11	11	11	11	11	10	10
GAC	16	16	16	16	16	16	16	16	17	17
GAA	13	13	13	13	13	13	13	13	13	13
GAG	10	10	10	10	10	10	10	10	10	10
TGT	4	4	4	4	4	4	4	4	4	4
TGC	1	1	1	1	1	1	1	1	1	1
TGA	4	4	4	4	4	4	4	4	4	4
TGG	12	12	12	12	12	12	12	12	12	12
CGT	0	0	0	0	0	0	0	0	0	0
CGC	1	1	1	1	1	1	1	1	1	1
CGA	1	1	1	1	1	1	1	1	1	1
CGG	0	0	0	0	0	0	0	0	0	0
ACT	5	5	5	5	5	5	5	5	5	5
AGC	3	3	3	3	3	3	3	3	3	3
AGA	7	7	7	7	7	7	7	7	7	7
AGG	4	4	4	4	4	4	4	4	4	4
GGT	22	22	22	22	22	22	22	22	22	22
GGC	7	7	7	7	7	7	7	7	7	7
GGA	7	7	7	7	7	7	7	7	7	7
GGG	1	1	1	1	1	1	1	1	1	1
TOTAL CODON	406	406	406	406	406	406	406	406	406	406

The treated 2nd larval instar with LC_{25} of Cascade showed two mutations after 24&48 hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.194)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, **Fig.(50)**. Therefore according to these two mutations the encoded amino acids showed two mutations. The first one involved the substitution of the amino acid **I (No.65)** with **T**, the second involved the substitution of the amino acid **L (No.156)** with **F**, **Fig.(51)**.

Exposed 2nd larval instar to LC_{25} of Match showed two mutations after 24&48 hrs post treatment. The first one involved the substitution of the nitrogenous base **G(No.468)** with **C,** the second mutation involved the substitution of the nitrogenous base **T (No.701)** with **C Fig.(54) .** Therefore according to these two mutations the encoded amino acids showed two mutations. The first one involved the substitution of the amino acid **L (No.156)** with **F**, the second involved the substitution of the amino acid **I (No.234)** with **T**, **Fig.(55)**.

The treated 2nd larval instar with LC_{25} of Mimic showed two mutations after 24&48 hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.384)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, **Fig.(56)**. Therefore according to these two mutations the encoded amino acids showed one mutation, involved the substitution of the amino acid **L (No.156)** with **F**, **Fig.(57)**.

The treated 2nd larval instar with LC_{50} and LC_{90} of Cascade showed three mutations after 24&48 hrs post treatment. The first one involved the substitution of the nitrogenous base **T (No.194)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, the third mutation involved the substitution of the nitrogenous base **G (No.730)** with **C, Fig.(58)**. Therefore according to these three mutations the encoded amino acids showed three mutations. The first one involved the substitution of the amino acid **I (No.65)** with **T**, the second involved the substitution of the amino acid **L (No.156)** with **F**, the third mutation involved the substitution of the amino acid **G (No.244)** with **R Fig.(59)**.

The treated 2nd larval instar with LC_{50} and LC_{90} of Match showed three mutations after 24&48 hrs post treatment. The first one involved the substitution of the nitrogenous base **G (No.260)** with **C**, the second mutation involved the substitution of the nitrogenous base **G (No.468)** with **C**, the third mutation involved the substitution of the nitrogenous base **T (No.701)** with **C, Fig.(60)**. Therefore according to these three mutations the encoded amino acids showed three mutations. The first one involved the substitution of the amino acid **R (No.87)** with **T**, the second involved the substitution of the amino acid **L (No.156)** with **F**, the third mutation involved the substitution of the amino acid **I (No.234)** with **T Fig.(61)**.

Accordingly, the following parameters have been changed**,** total codones numbers which represented in **table (37)** and amino acid numbers which represented in **table (38).**

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGAT►CCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCA
CTGACGAAGTGAAGAGACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACAT
TGATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATG
AAGGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTG►CACATCTGCTATCAGCTCTGG
TTGGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAG
GGAGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTG
ACTTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGATCGTTGTTGGTGTTGCAATGTATGGAAGAG►CGT
TGGACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATG
GTGTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACC
GTATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGG
AACAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCG
GTAACAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATG
TCTCAAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (58): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Cascade after 24&24 hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKI►TLPSVGGWTLADPFFF
FTDEVKRHRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYEL►FTSAI
SSGWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKIVVGVAMYG
RG►RWTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKA
KYVRNNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KK
SL
```

Fig. (59): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Cascade after 24&24 hrs.

```
GTAAAGGAGATTGAAGGCAGTTTCCAAGCTCTTCAGAGATCTTGCAGCGGCAGGGAAGACTTCAAAGTATCTATCCACGATC
CTTGGGCTGCTCTTCAGAAACCTCAAAAAGGTTTGTCATCCTGGAACGAGCCTTACAAGGGTAACTTCGGCCAATTGATGAT
GTTGAAACAAGCAAGACCAGATCTGAAGATCTTGCCTTCTGTTGGTGGATGGACCCTGGCTGATCCGTTCTTCTTTTTCACT
GACGAAGTGAAGAG►CACACCGCTTCGTTGCTTCAGTCAAGGACTTCCTTGAAACCTGGAAGTTCTTTGATGGTGTTGACAT
TGATTGGGAATTCCCTGGTGGTAAAGGAGCCAACCCTGATCTCGGCGCCCCTGAAGATGGTCACATCTACGTCCAGCTGATG
AAGGAACTCAGAGAGATGTTGAACGAGCTTTCTGCCAAGACTGGTAAGAAGTACGAATTG►CACATCTGCTATCAGCTCTGG
TTGGGACAAGATTCAGGTTGTGGACTACAAAGAAGCTCAGCAGTACATGGACCACATCTTCTTGATGAGTTATGACTTCAAG
GGAGCTTGGTCTAACGACACTCTTGGCCACCAGGCTGGTTTGTACGCCCCAGCATGGAACCCCAAGGAGACTTACACAACTG
ACTTTGGTGTCAAATTCCTGTTAGCACAAGGTGTCAGCCCCAAGAAGAT►CCGTTGTTGGTGTTGCAATGTATGGAAGAGGT
TGGACTGGAGTCAACGGCTACAAAGATGGTAATCCGTTCACTGGTGTAGCTACTGGTCCAGTTAAAGGAACTTGGCAGGATG
GTGTGGTAGACTACAGAGAGATTGCCAATGAAATTGCTCAGGGCAAGTGGGAGTATCACTATGATAAGGTGGCTCAGGCACC
GTATGTGTTCAGGAAGGAGACTGGAGACCTTATTACGTATGATGACGCGAGATCGACAATTGAGAAAGCTAAATATGTGAGG
AACAATAAGTTAGGTGGGTTGTTCGCTTGGGAGGTGGACGCGGACAATGGTGACATATTAAATGCCATGAATATGGGTCTCG
GTAACAATGCGTGAAGGGAAACTAGTGCGAGTGTTGTGTGTAAATTGTGATCGTAATGATAAGTTTAGGTTTAGGCTGAATG
TCTCAAATTTTTATTTACAAAAATTGTTGAACGACTGAATCTGTGTAATTGTAAGTTAATATGAAAAAAAAGTTTA
```

Fig. (60): The expected sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Match after 24&48 hrs.

```
VKEIEGSFQALQRSCSGREDFKVSIHDPWAALQKPQKGLSSWNEPYKGNFGQLMMLKQARPDLKILPSVGGWTLADPFFFFT
DEVKR►THRFVASVKDFLETWKFFDGVDIDWEFPGGKGANPDLGAPEDGHIYVQLMKELREMLNELSAKTGKKYEL►FTSAI
SSGWDKIQVVDYKEAQQYMDHIFLMSYDFKGAWSNDTLGHQAGLYAPAWNPKETYTTDFGVKFLLAQGVSPKKI►TVVGVAM
YGRGWTGVNGYKDGNPFTGVATGPVKGTWQDGVVDYREIANEIAQGKWEYHYDKVAQAPYVFRKETGDLITYDDARSTIEKA
KYVRNNKLGGLFAWEVDADNGDILNAMNMGLGNNA*RETSASVVCKL*S***V*V*AECLKFLFTKIVERLNLCNCKLI*KK
SL
```

Fig. (61): The expected protein sequence of the mutated vira-like chitinase gene of the treated 2nd larval instar with LC_{50} and LC_{90} of Match after 24&48 h

Table (37): change in total number of codons after 24&48hrs of treatment.

codons	Number of codons									
	NOR.	CAS. LC_{25}	CAS. LC_{50}	CAS. LC_{90}	MAT. LC_{25}	MAT. LC_{50}	MAT. LC_{90}	MIM. LC_{25}	MIM. LC_{50}	MIM. LC_{90}
TTT	5	5	5	5	5	5	5	5	5	5
TTC	16	17	17	17	17	17	17	17	17	17
TTA	6	6	6	6	6	6	6	6	6	6
TTG	10	9	9	9	9	9	9	9	9	9
CTT	6	6	6	6	6	6	6	6	6	6
CTC	4	4	4	4	4	4	4	4	4	4
CTA	0	0	0	0	0	0	0	0	0	0
CTG	6	6	6	6	6	6	6	6	6	6
ATT	8	8	8	8	8	8	8	8	8	8
ATC	6	5	5	5	5	5	5	6	6	6
ATA	2	2	2	2	2	2	2	2	2	2
ATG	9	9	9	9	9	9	9	9	9	9
GTT	12	12	12	12	12	12	12	12	12	12
GTC	5	5	5	5	5	5	5	5	5	5
GTA	4	4	4	4	4	4	4	4	4	4
GTG	8	8	8	8	8	8	8	8	8	8
TCT	7	7	7	7	7	7	7	7	7	7
TCC	1	1	1	1	1	1	1	1	1	1
TCA	2	2	2	2	2	2	2	2	2	2
TCG	2	2	2	2	2	2	2	2	2	2
CCT	7	7	7	7	7	7	7	7	7	7
CCC	2	2	2	2	2	2	2	2	2	2
CCA	3	3	3	3	3	3	3	3	3	3
CCG	3	3	3	3	3	3	3	3	3	3
ACT	11	11	11	11	11	11	11	11	11	11
ACC	2	3	3	3	3	3	3	2	2	2
ACA	4	4	4	4	4	5	5	4	4	4
ACG	1	1	1	1	1	1	1	1	1	1
GCT	15	15	15	15	15	15	15	15	15	15
GCC	6	6	6	6	6	6	6	6	6	6
GCA	5	5	5	5	5	5	5	5	5	5
GCG	4	4	4	4	4	4	4	4	4	4

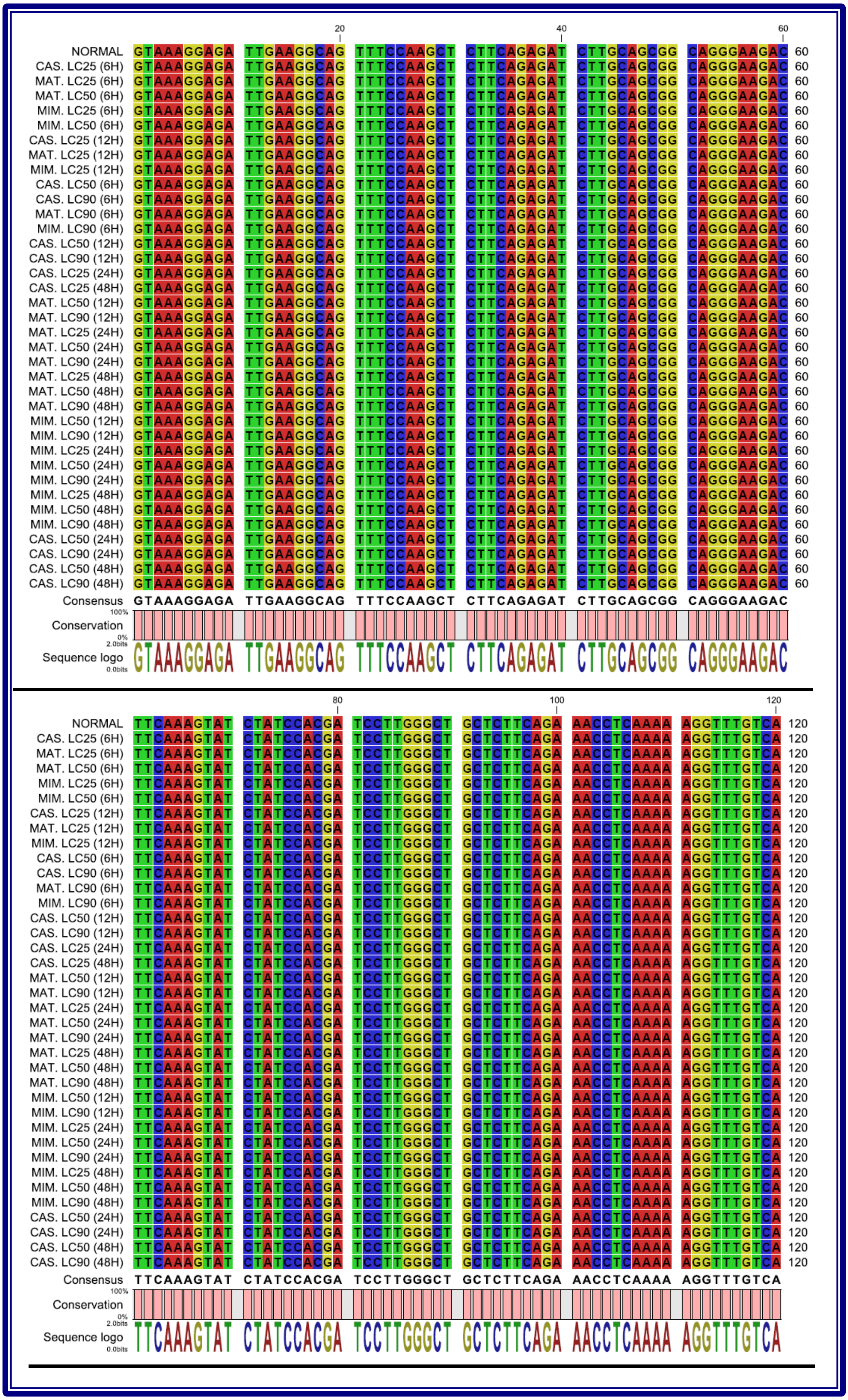
Continued table (37)

codons	Number of codons									
	NOR.	CAS. LC$_{25}$	CAS. LC$_{50}$	CAS. LC$_{90}$	MAT. LC$_{25}$	MAT. LC$_{50}$	MAT. LC$_{90}$	MIM. LC$_{25}$	CIM. LC$_{50}$	MIM. LC$_{90}$
TAT	7	7	7	7	7	7	7	7	7	7
TAC	9	9	9	9	9	9	9	9	9	9
TAA	2	2	2	2	2	2	2	2	2	2
TAG	2	2	2	2	2	2	2	2	2	2
CAT	0	0	0	0	0	0	0	0	0	0
CAC	6	6	6	6	6	6	6	6	6	6
CAA	5	5	5	5	5	5	5	5	5	5
CAG	10	10	10	10	10	10	10	10	10	10
AAT	9	9	9	9	9	9	9	9	9	9
AAC	9	9	9	9	9	9	9	9	9	9
AAA	16	16	16	16	16	16	16	16	16	16
AAG	20	20	20	20	20	20	20	20	20	20
GAT	11	11	11	11	11	11	11	10	10	10
GAC	16	16	16	16	16	16	16	17	17	17
GAA	13	13	13	13	13	13	13	13	13	13
GAG	10	10	10	10	10	10	10	10	10	10
TGT	4	4	4	4	4	4	4	4	4	4
TGC	1	1	1	1	1	1	1	1	1	1
TGA	4	4	4	4	4	4	4	4	4	4
TGG	12	12	12	12	12	12	12	12	12	12
CGT	0	0	1	1	1	1	1	0	0	0
CGC	1	1	1	1	1	1	1	1	1	1
CGA	1	1	1	1	1	1	1	1	1	1
CGG	0	0	0	0	0	0	0	0	0	0
ACT	5	5	5	5	5	5	5	5	5	5
AGC	3	3	3	3	3	3	3	3	3	3
AGA	7	7	7	7	7	6	6	7	7	7
AGG	4	4	4	4	4	4	4	4	4	4
GGT	22	22	21	21	21	21	21	22	22	22
GGC	7	7	7	7	7	7	7	7	7	7
GGA	7	7	7	7	7	7	7	7	7	7
GGG	1	1	1	1	1	1	1	1	1	1
Total Codons	406	406	406	406	406	406	406	406	406	406

4.5.2 Vira-like chitinase gene alignment:-

The alignments of the vira-like chitinase gene sequences and the encoded amino acids for the studied groups were obtained by CLC Main Workbench program (version 5.5) and shown in **figures (62&63)** respectively.

The consensus sequence and sequence logo are represented in the alignment, taking into account that the program does not consider the 1st group **(Normal)** as a reference one.



```
                                           20                                          40                                          60
                                            |                                           |                                           |
NORMAL          GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC25 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MAT. LC25 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MAT. LC50 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MIM. LC25 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MIM. LC50 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC25 (12H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MAT. LC25 (12H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
MIM. LC25 (12H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC50 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC90 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
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MIM. LC90 (6H)  GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
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CAS. LC90 (12H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC25 (24H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
CAS. LC25 (48H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
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MAT. LC90 (12H) GTAAAGGAGA TTGAAGGCAG TTTCCAAGCT CTTCAGAGAT CTTGCAGCGG CAGGGAAGAC 60
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Conservation
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        0.0bits

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MAT. LC25 (6H)  TTCAAAGTAT CTATCCACGA TCCTTGGGCT GCTCTTCAGA AACCTCAAAA AGGTTTGTCA 120
MAT. LC50 (6H)  TTCAAAGTAT CTATCCACGA TCCTTGGGCT GCTCTTCAGA AACCTCAAAA AGGTTTGTCA 120
MIM. LC25 (6H)  TTCAAAGTAT CTATCCACGA TCCTTGGGCT GCTCTTCAGA AACCTCAAAA AGGTTTGTCA 120
MIM. LC50 (6H)  TTCAAAGTAT CTATCCACGA TCCTTGGGCT GCTCTTCAGA AACCTCAAAA AGGTTTGTCA 120
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Conservation
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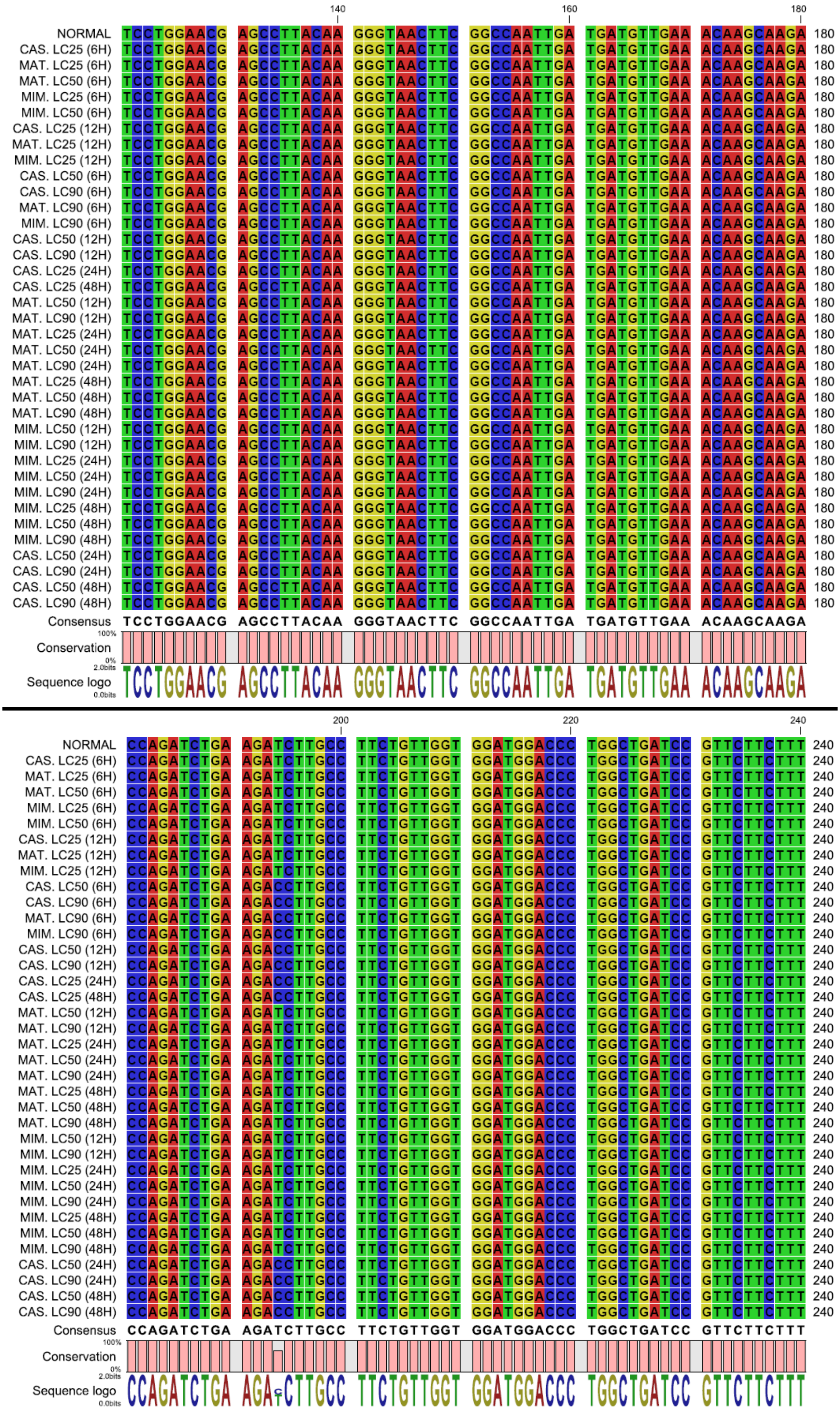

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MAT. LC25 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC50 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC25 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC50 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC25 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC25 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC25 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC50 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC90 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC90 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC90 (6H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC50 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC90 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC25 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC25 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC50 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC90 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
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MAT. LC50 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MAT. LC90 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
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MAT. LC90 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
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MIM. LC90 (12H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC25 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC50 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
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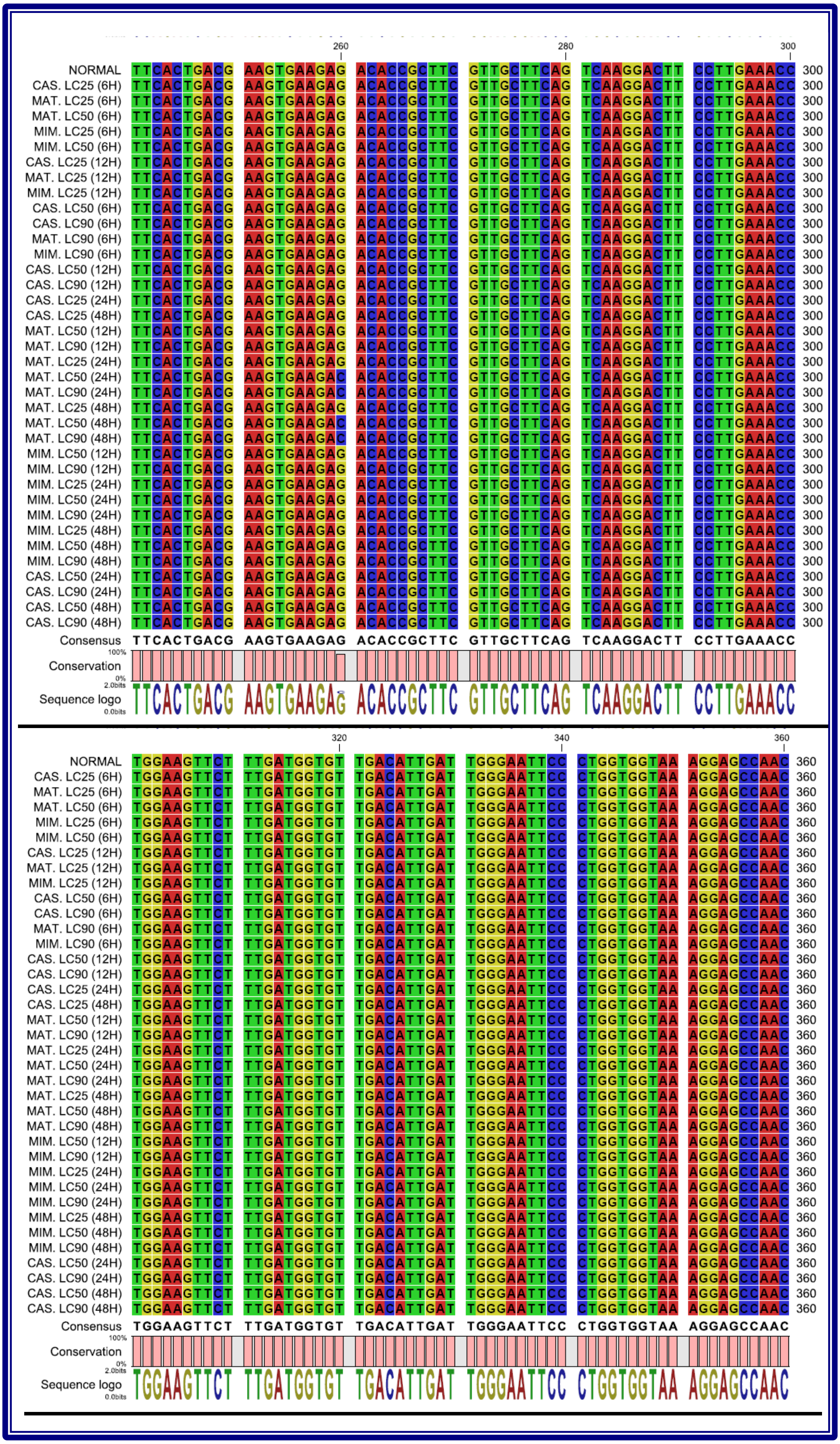
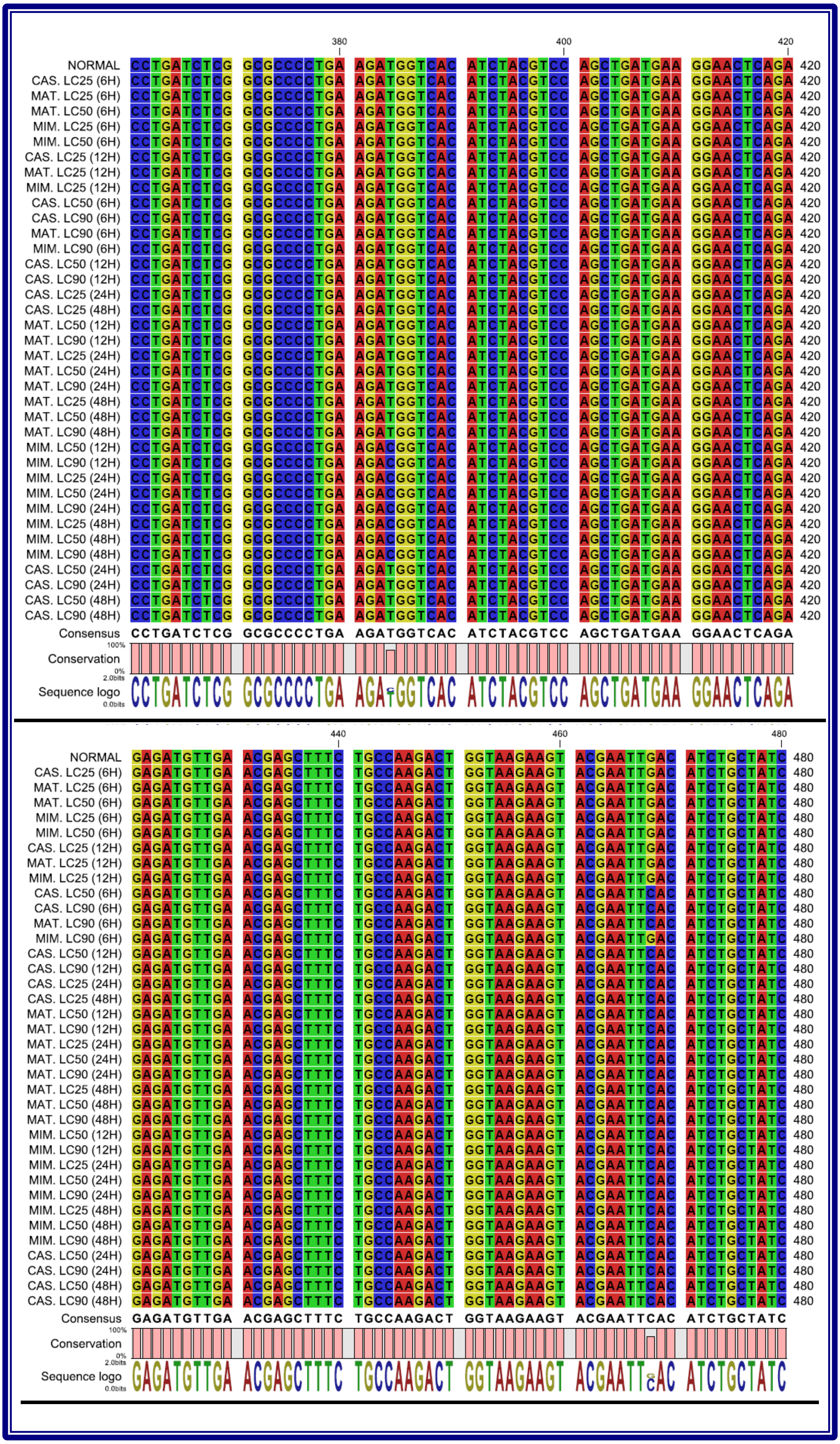
MIM. LC25 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC50 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
MIM. LC90 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC50 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC90 (24H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC50 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
CAS. LC90 (48H)	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	180
Consensus	TCCTGGAACG	AGCCTTACAA	GGGTAACTTC	GGCCAATTGA	TGATGTTGAA	ACAAGCAAGA	

		200		220		240	
NORMAL	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC25 (6H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC25 (6H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC50 (6H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC25 (6H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC50 (6H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC25 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC25 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC25 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC50 (6H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC90 (6H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC90 (6H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC90 (6H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC50 (12H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC90 (12H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC25 (24H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC25 (48H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC50 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC90 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC25 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC50 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC90 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC25 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC50 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MAT. LC90 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC50 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC90 (12H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC25 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC50 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC90 (24H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC25 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC50 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
MIM. LC90 (48H)	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC50 (24H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC90 (24H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC50 (48H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
CAS. LC90 (48H)	CCAGATCTGA	AGACCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	240
Consensus	CCAGATCTGA	AGATCTTGCC	TTCTGTTGGT	GGATGGACCC	TGGCTGATCC	GTTCTTCTTT	



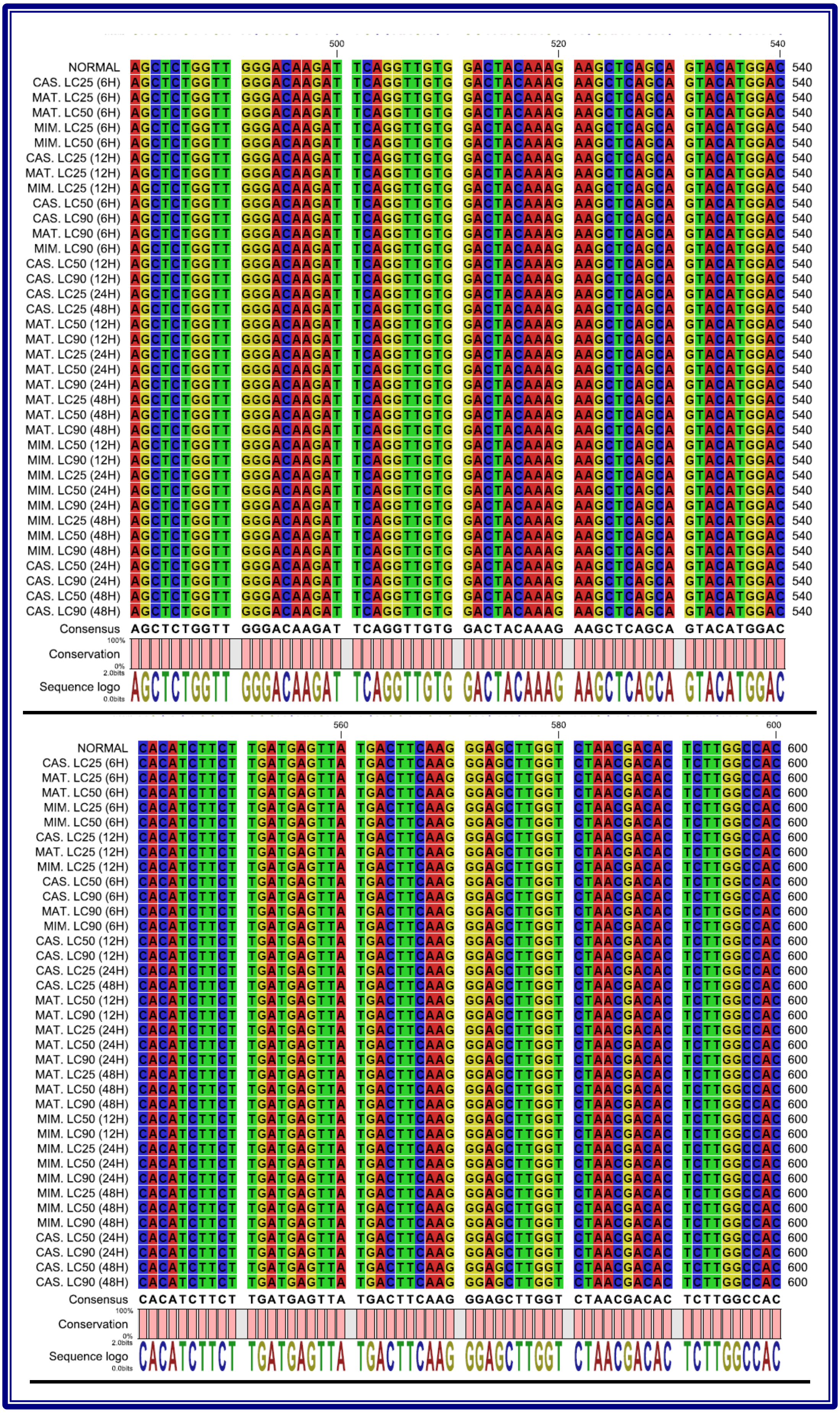
		260		280		300	
NORMAL	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC25 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC25 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC50 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC25 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC50 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC25 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC25 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC25 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC50 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC90 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC90 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC90 (6H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC50 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC90 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC25 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC25 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC50 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC90 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC25 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC50 (24H)	TTCACTGACG	AAGTGAAGAC	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC90 (24H)	TTCACTGACG	AAGTGAAGAC	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC25 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC50 (48H)	TTCACTGACG	AAGTGAAGAC	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MAT. LC90 (48H)	TTCACTGACG	AAGTGAAGAC	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC50 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC90 (12H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC25 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC50 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC90 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC25 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC50 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
MIM. LC90 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC50 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC90 (24H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC50 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
CAS. LC90 (48H)	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	300
Consensus	TTCACTGACG	AAGTGAAGAG	ACACCGCTTC	GTTGCTTCAG	TCAAGGACTT	CCTTGAAACC	

		320		340		360	
NORMAL	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC25 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC25 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC50 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC25 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC50 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC25 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC25 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC25 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC50 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC90 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC90 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC90 (6H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC50 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC90 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC25 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC25 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC50 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC90 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC25 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC50 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC90 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC25 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC50 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MAT. LC90 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC50 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC90 (12H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC25 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC50 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC90 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC25 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC50 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
MIM. LC90 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC50 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC90 (24H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC50 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
CAS. LC90 (48H)	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	360
Consensus	TGGAAGTTCT	TTGATGGTGT	TGACATTGAT	TGGGAATTCC	CTGGTGGTAA	AGGAGCCAAC	



		380		400		420	
NORMAL	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC25 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC25 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC50 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC25 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC50 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC25 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC25 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC25 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC50 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC90 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC90 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC90 (6H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC50 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC90 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC25 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC25 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC50 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC90 (12H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC25 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC50 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC90 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC25 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC50 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MAT. LC90 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC50 (12H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC90 (12H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC25 (24H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC50 (24H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC90 (24H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC25 (48H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC50 (48H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
MIM. LC90 (48H)	CCTGATCTCG	GCGCCCCTGA	AGACGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC50 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC90 (24H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC50 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
CAS. LC90 (48H)	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	420
Consensus	CCTGATCTCG	GCGCCCCTGA	AGATGGTCAC	ATCTACGTCC	AGCTGATGAA	GGAACTCAGA	

		440		460		480	
NORMAL	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
CAS. LC25 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MAT. LC25 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MAT. LC50 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MIM. LC25 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MIM. LC50 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
CAS. LC25 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MAT. LC25 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
MIM. LC25 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
CAS. LC50 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC90 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC90 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC90 (6H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTGAC	ATCTGCTATC	480
CAS. LC50 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC90 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC25 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC25 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC50 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC90 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC25 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC50 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC90 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC25 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC50 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MAT. LC90 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC50 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC90 (12H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC25 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC50 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC90 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC25 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC50 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
MIM. LC90 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC50 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC90 (24H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC50 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
CAS. LC90 (48H)	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	480
Consensus	GAGATGTTGA	ACGAGCTTTC	TGCCAAGACT	GGTAAGAAGT	ACGAATTCAC	ATCTGCTATC	



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CAS. LC25 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC25 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC50 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MIM. LC25 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MIM. LC50 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC25 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC25 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MIM. LC25 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC50 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC90 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC90 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MIM. LC90 (6H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC50 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC90 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC25 (24H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC25 (48H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC50 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC90 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC25 (24H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
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MAT. LC50 (48H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MAT. LC90 (48H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
MIM. LC50 (12H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
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CAS. LC50 (48H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
CAS. LC90 (48H)	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	540
Consensus	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	
Sequence logo	AGCTCTGGTT	GGGACAAGAT	TCAGGTTGTG	GACTACAAAG	AAGCTCAGCA	GTACATGGAC	

NORMAL	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC25 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MAT. LC25 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MAT. LC50 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MIM. LC25 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MIM. LC50 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC25 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MAT. LC25 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MIM. LC25 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC50 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC90 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MAT. LC90 (6H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
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CAS. LC50 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC90 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC25 (24H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC25 (48H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
MAT. LC50 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
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MIM. LC50 (12H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
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MIM. LC25 (48H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
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MIM. LC90 (48H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
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CAS. LC90 (24H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC50 (48H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
CAS. LC90 (48H)	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	600
Consensus	CACATCTTCT	TGATGAGTTA	TGACTTCAAG	GGAGCTTGGT	CTAACGACAC	TCTTGGCCAC	
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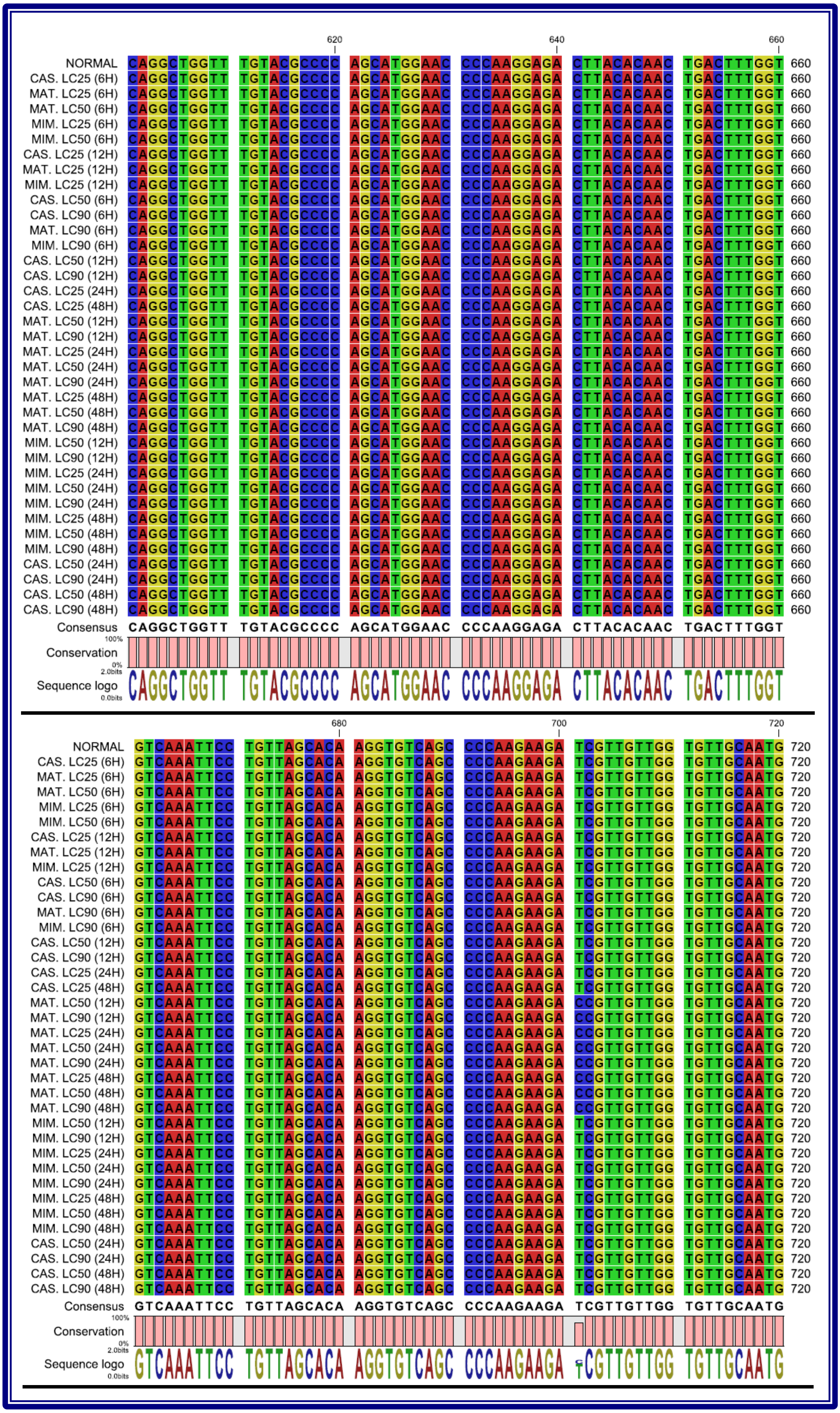

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MAT. LC50 (6H)	CAGGCTGGTT	TGTACGCCCC	AGCATGGAAC	CCCAAGGAGA	CTTACACAAC	TGACTTTGGT	660
MIM. LC25 (6H)	CAGGCTGGTT	TGTACGCCCC	AGCATGGAAC	CCCAAGGAGA	CTTACACAAC	TGACTTTGGT	660
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CAS. LC50 (48H)	CAGGCTGGTT	TGTACGCCCC	AGCATGGAAC	CCCAAGGAGA	CTTACACAAC	TGACTTTGGT	660
CAS. LC90 (48H)	CAGGCTGGTT	TGTACGCCCC	AGCATGGAAC	CCCAAGGAGA	CTTACACAAC	TGACTTTGGT	660
Consensus	CAGGCTGGTT	TGTACGCCCC	AGCATGGAAC	CCCAAGGAGA	CTTACACAAC	TGACTTTGGT	
Conservation (100% / 0%)							
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NORMAL	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
CAS. LC25 (6H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
MAT. LC25 (6H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
MAT. LC50 (6H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
MIM. LC25 (6H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
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CAS. LC50 (48H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
CAS. LC90 (48H)	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	720
Consensus	GTCAAATTCC	TGTTAGCACA	AGGTGTCAGC	CCCAAGAAGA	TCGTTGTTGG	TGTTGCAATG	
Conservation (100% / 0%)							
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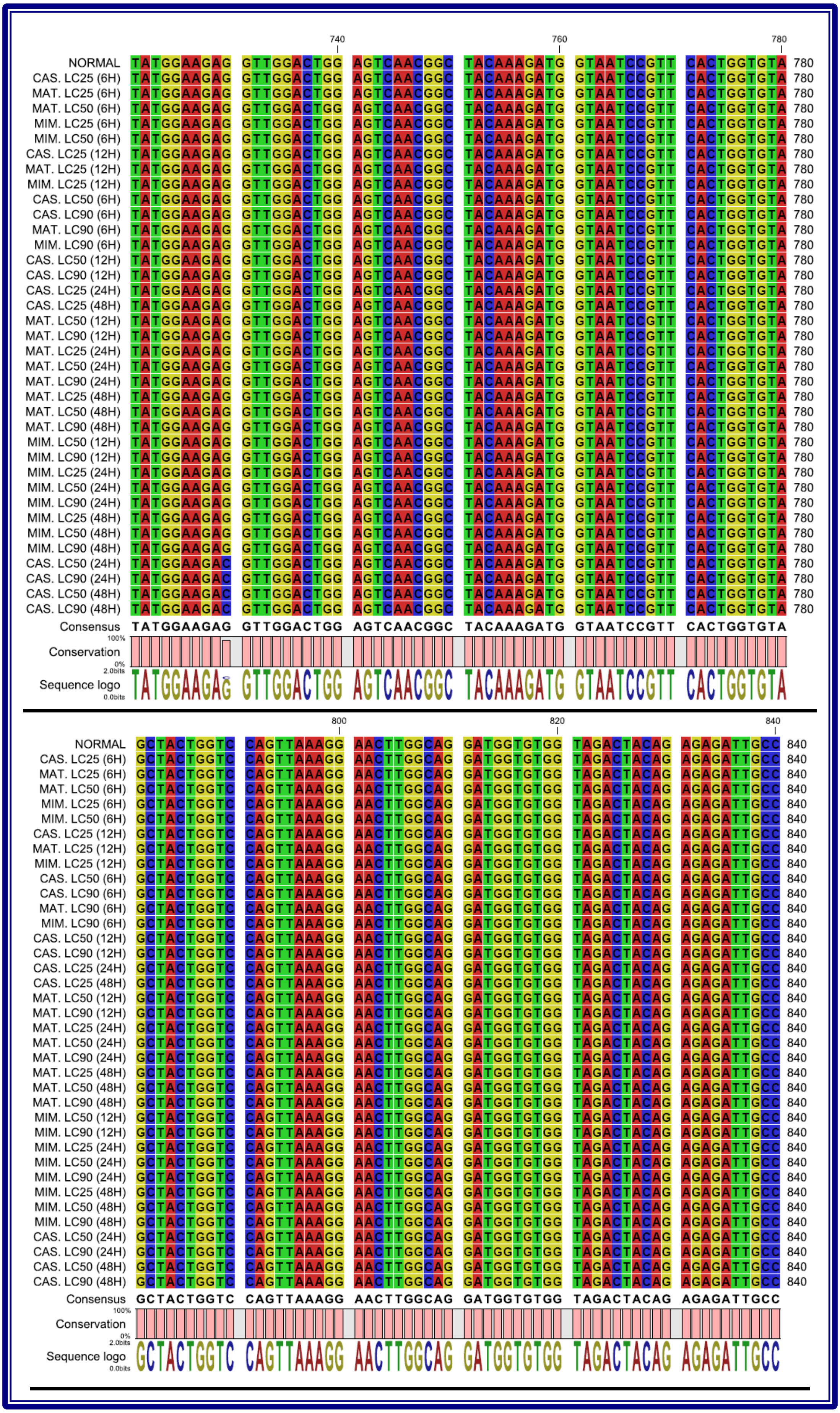

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NORMAL	TATGGAAGAG	GTTGGACTGG	AGTCAACGGC	TACAAAGATG	GTAATCCGTT	CACTGGTGTA	780
CAS. LC25 (6H)	TATGGAAGAG	GTTGGACTGG	AGTCAACGGC	TACAAAGATG	GTAATCCGTT	CACTGGTGTA	780
MAT. LC25 (6H)	TATGGAAGAG	GTTGGACTGG	AGTCAACGGC	TACAAAGATG	GTAATCCGTT	CACTGGTGTA	780
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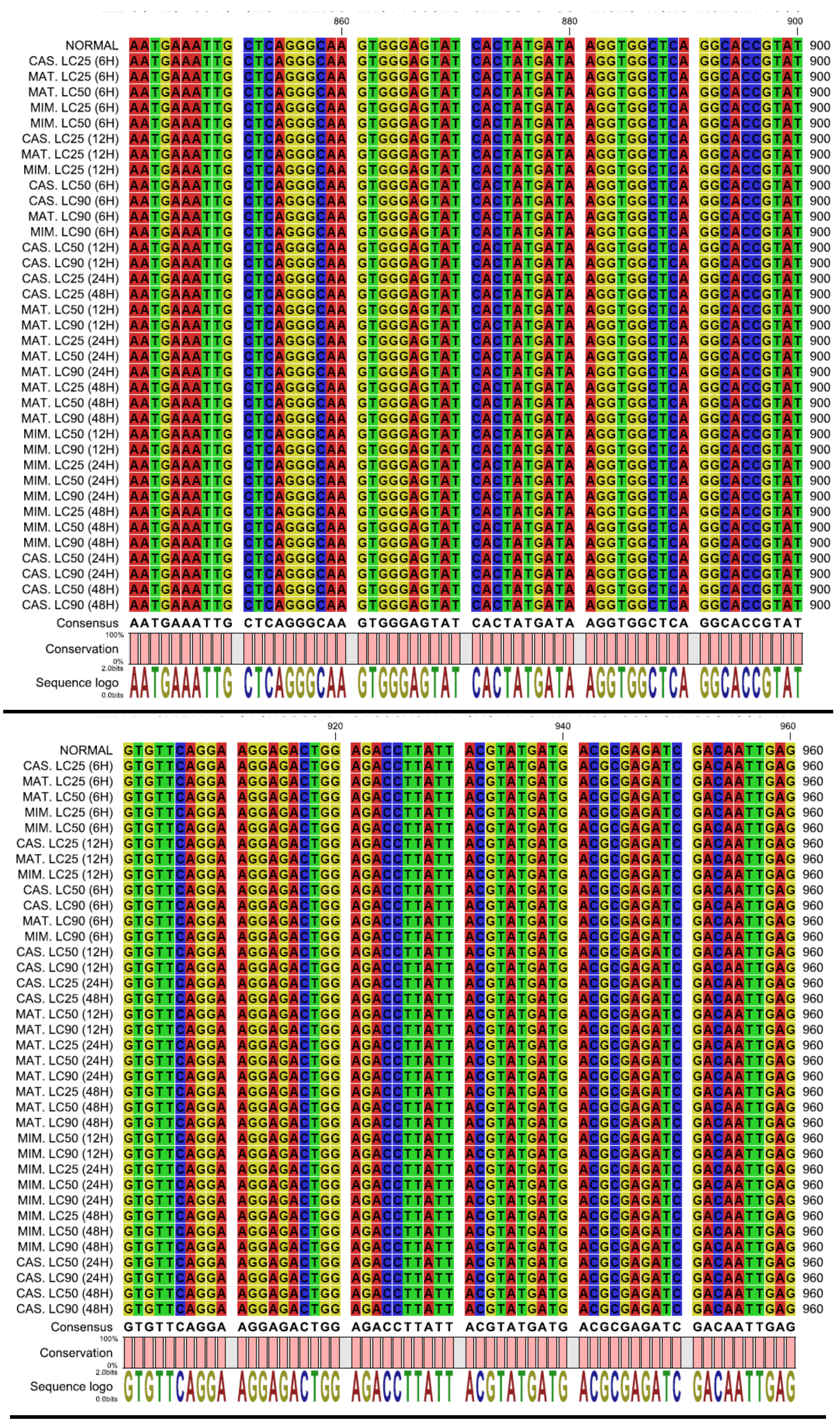

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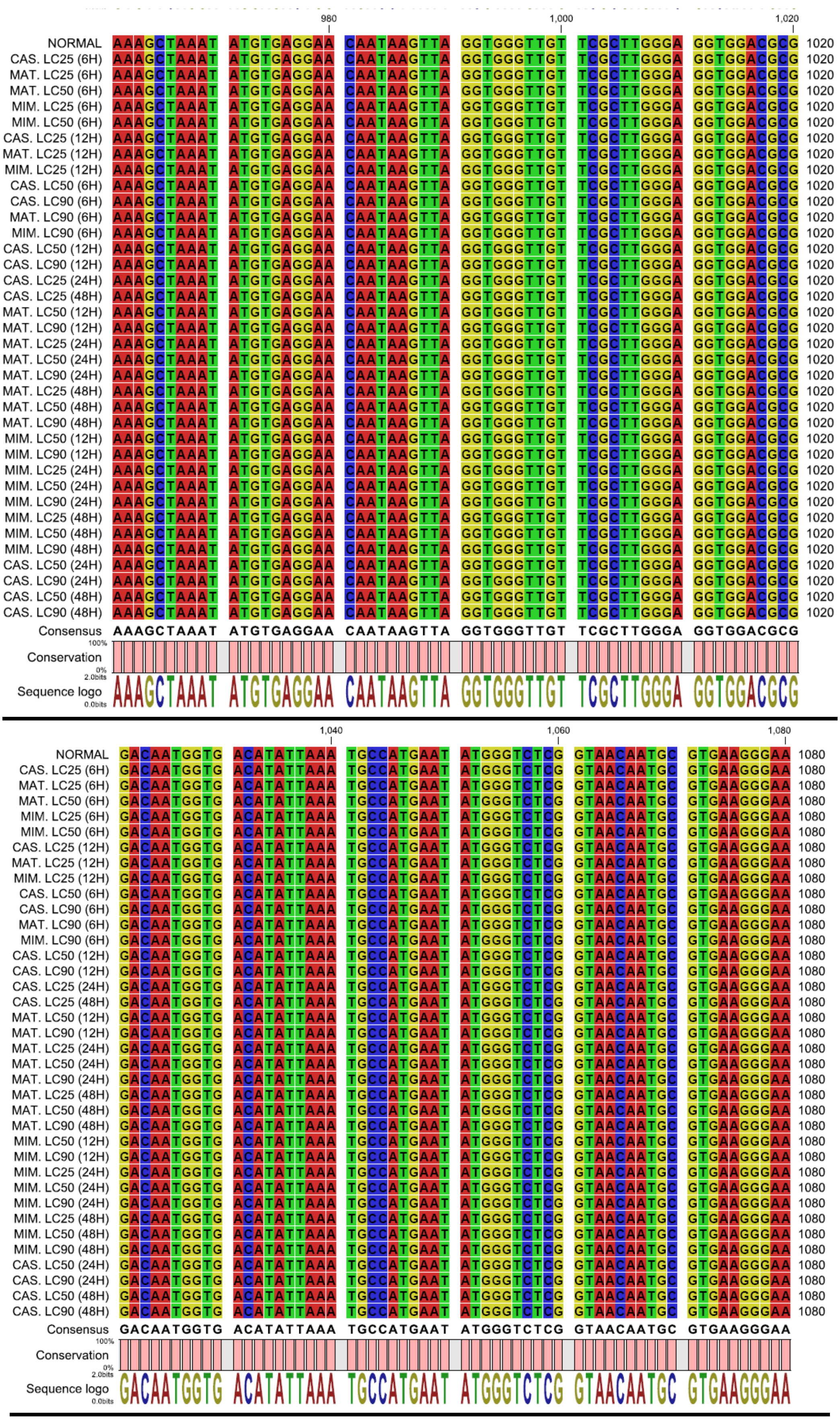

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CAS. LC90 (48H)   AAAGCTAAAT ATGTGAGGAA CAATAAGTTA GGTGGGTTGT TCGCTTGGGA GGTGGACGCG 1020
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MAT. LC25 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MAT. LC50 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MIM. LC25 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MIM. LC50 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
CAS. LC25 (12H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MAT. LC25 (12H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MIM. LC25 (12H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
CAS. LC50 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
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MIM. LC90 (6H)    GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
CAS. LC50 (12H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
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CAS. LC25 (24H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
CAS. LC25 (48H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
MAT. LC50 (12H)   GACAATGGTG ACATATTAAA TGCCATGAAT ATGGGTCTCG GTAACAATGC GTGAAGGGAA 1080
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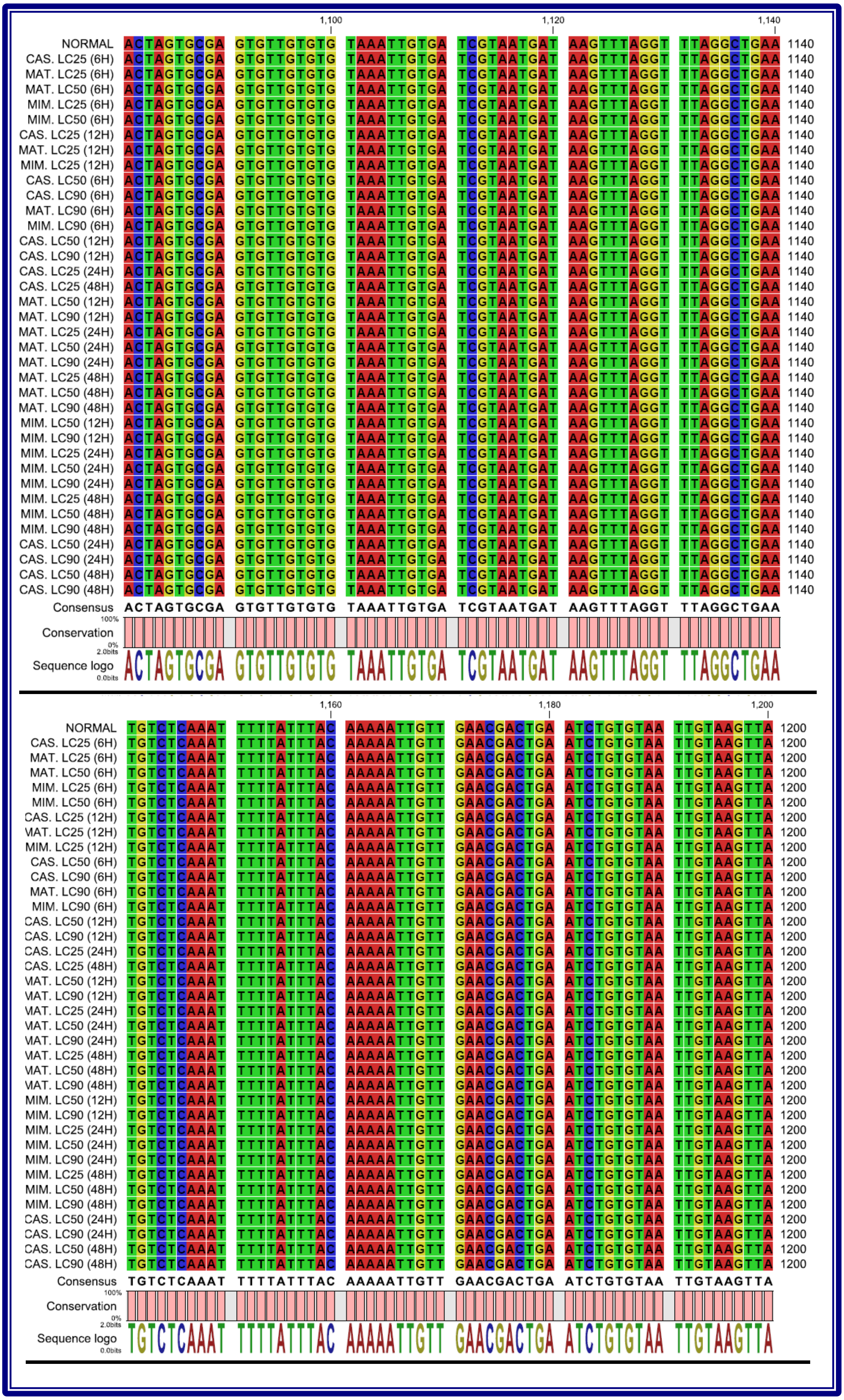

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MAT. LC50 (6H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
MIM. LC25 (6H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
MIM. LC50 (6H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
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CAS. LC50 (6H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
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MIM. LC90 (6H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
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MAT. LC25 (48H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
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MIM. LC90 (24H)	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	1140
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Consensus	ACTAGTGCGA	GTGTTGTGTG	TAAATTGTGA	TCGTAATGAT	AAGTTTAGGT	TTAGGCTGAA	
Conservation							
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MAT. LC25 (12H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
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CAS. LC25 (48H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
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MAT. LC90 (24H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
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CAS. LC50 (24H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
CAS. LC90 (24H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
CAS. LC50 (48H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
CAS. LC90 (48H)	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	1200
Consensus	TGTCTCAAAT	TTTTATTTAC	AAAAATTGTT	GAACGACTGA	ATCTGTGTAA	TTGTAAGTTA	
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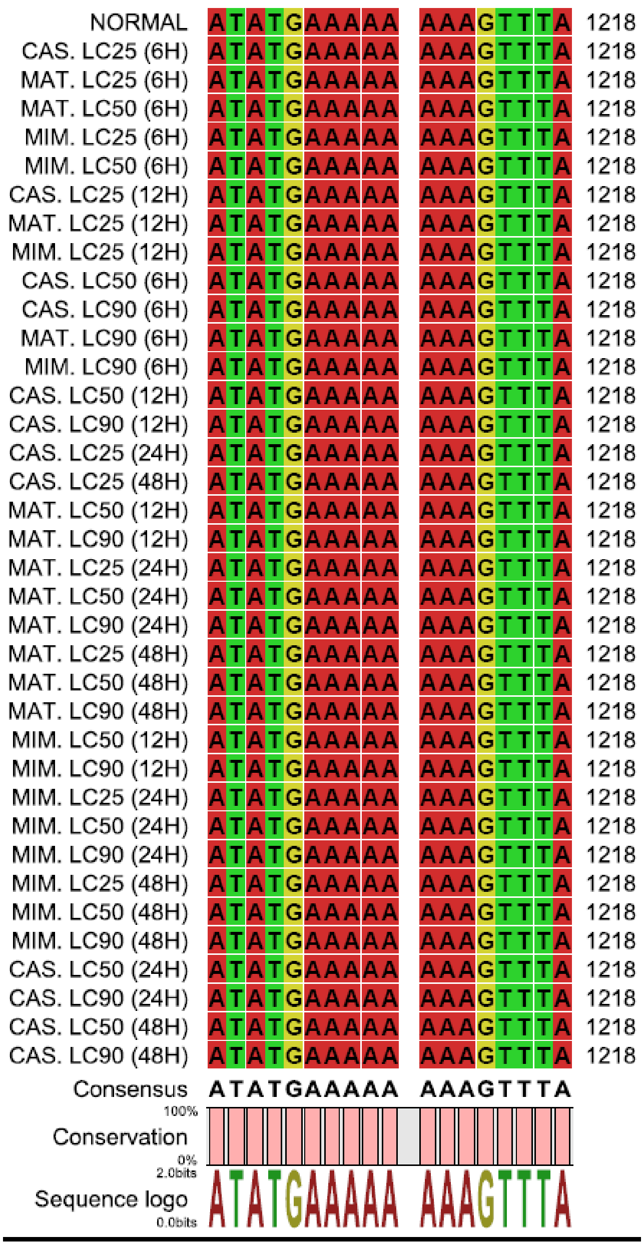

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MIM. LC25 (12H)	ATATGAAAAA	AAAGTTTA	1218
CAS. LC50 (6H)	ATATGAAAAA	AAAGTTTA	1218
CAS. LC90 (6H)	ATATGAAAAA	AAAGTTTA	1218
MAT. LC90 (6H)	ATATGAAAAA	AAAGTTTA	1218
MIM. LC90 (6H)	ATATGAAAAA	AAAGTTTA	1218
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MAT. LC50 (12H)	ATATGAAAAA	AAAGTTTA	1218
MAT. LC90 (12H)	ATATGAAAAA	AAAGTTTA	1218
MAT. LC25 (24H)	ATATGAAAAA	AAAGTTTA	1218
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MAT. LC90 (24H)	ATATGAAAAA	AAAGTTTA	1218
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MIM. LC25 (48H)	ATATGAAAAA	AAAGTTTA	1218
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MIM. LC90 (48H)	ATATGAAAAA	AAAGTTTA	1218
CAS. LC50 (24H)	ATATGAAAAA	AAAGTTTA	1218
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CAS. LC50 (48H)	ATATGAAAAA	AAAGTTTA	1218
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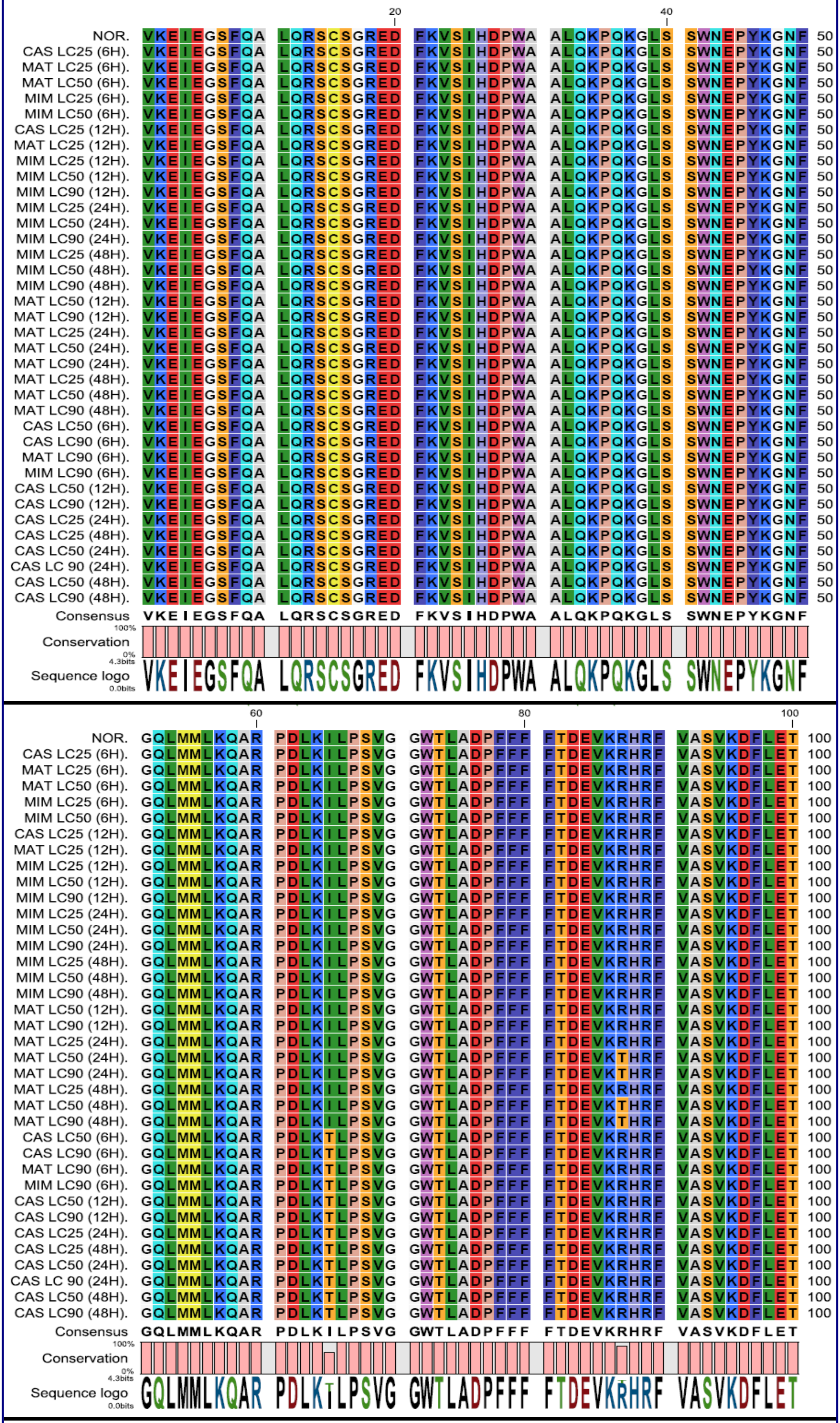
Fig. (62): Shows the alignment of the vira-like chitinas gene sequences for the studied groups.



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MAT LC50 (6H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
MIM LC25 (6H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
MIM LC50 (6H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
CAS LC25 (12H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MIM LC25 (24H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MIM LC90 (24H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
MIM LC25 (48H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MAT LC50 (12H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MAT LC90 (24H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
MAT LC25 (48H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MAT LC90 (48H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MAT LC90 (6H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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CAS LC50 (12H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
CAS LC90 (12H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
CAS LC25 (24H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
CAS LC25 (48H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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CAS LC90 (48H).	VKEIEGSFQA	LQRSCSGRED	FKVSIHDPWA	ALQKPQKGLS	SWNEPYKGNF	50
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MIM LC50 (6H).	GQLMMLKQAR	PDLKILPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
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MAT LC25 (12H).	GQLMMLKQAR	PDLKILPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
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MIM LC25 (48H).	GQLMMLKQAR	PDLKILPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
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CAS LC25 (24H).	GQLMMLKQAR	PDLKTLPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
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CAS LC 90 (24H).	GQLMMLKQAR	PDLKTLPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
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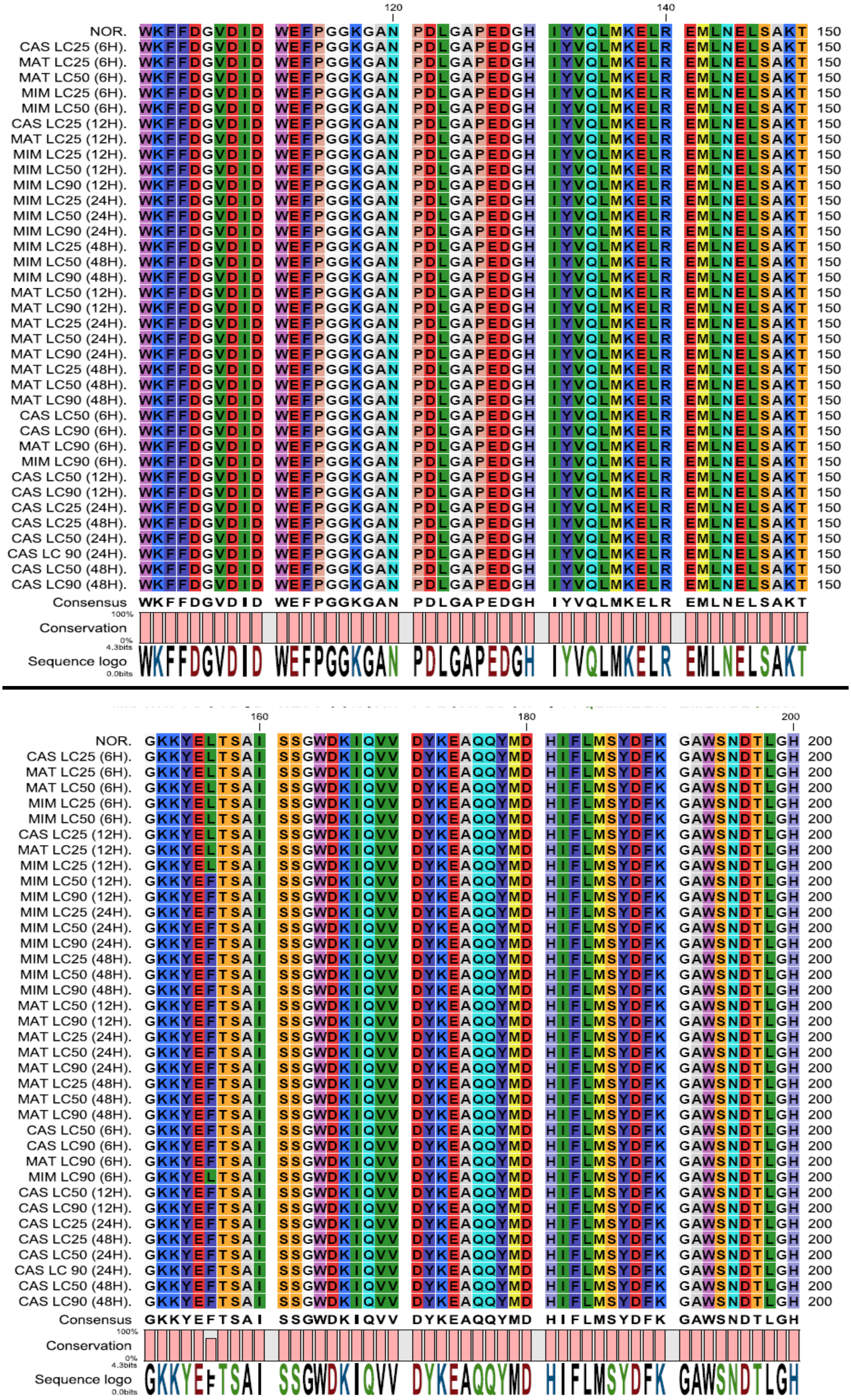
CAS LC90 (48H).	GQLMMLKQAR	PDLKTLPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	100
Consensus	GQLMMLKQAR	PDLKILPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	
Sequence logo	GQLMMLKQAR	PDLKTLPSVG	GWTLADPFFF	FTDEVKRHRF	VASVKDFLET	



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  CAS LC25 (6H). WKFFDGVDID WEFPGGKGAN PDLGAPEDGH IYVQLMKELR EMLNELSAKT 150
  MAT LC25 (6H). WKFFDGVDID WEFPGGKGAN PDLGAPEDGH IYVQLMKELR EMLNELSAKT 150
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 CAS LC90 (48H). WKFFDGVDID WEFPGGKGAN PDLGAPEDGH IYVQLMKELR EMLNELSAKT 150
       Consensus WKFFDGVDID WEFPGGKGAN PDLGAPEDGH IYVQLMKELR EMLNELSAKT
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                            |                     |                     |
            NOR. GKKYELTSAI SSGWDKIQVV DYKEAQQYMD HIFLMSYDFK GAWSNDTLGH 200
  CAS LC25 (6H). GKKYELTSAI SSGWDKIQVV DYKEAQQYMD HIFLMSYDFK GAWSNDTLGH 200
  MAT LC25 (6H). GKKYELTSAI SSGWDKIQVV DYKEAQQYMD HIFLMSYDFK GAWSNDTLGH 200
  MAT LC50 (6H). GKKYELTSAI SSGWDKIQVV DYKEAQQYMD HIFLMSYDFK GAWSNDTLGH 200
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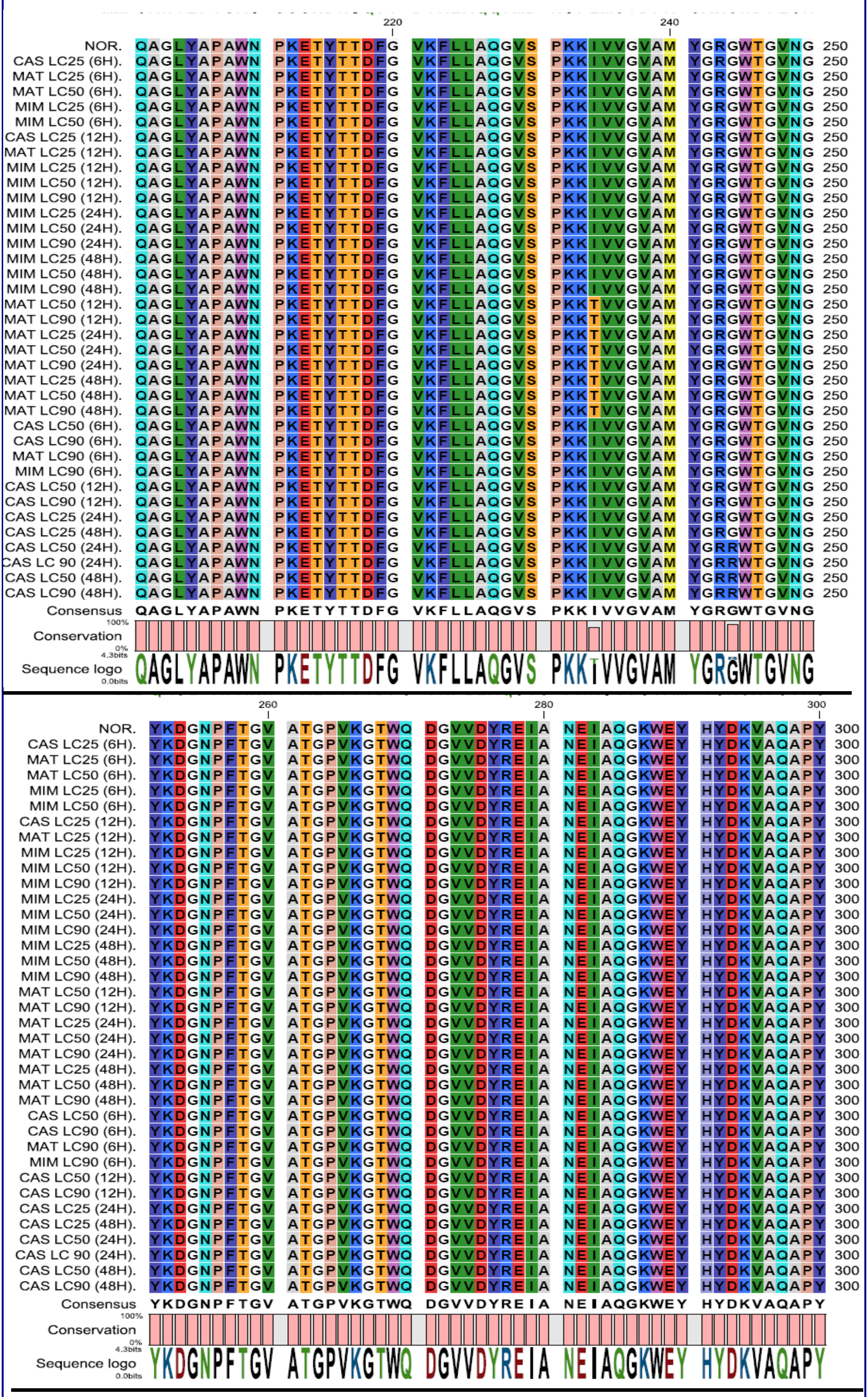

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MAT LC25 (6H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
MAT LC50 (6H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
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MIM LC50 (6H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
CAS LC25 (12H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
MAT LC25 (12H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
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MIM LC50 (48H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
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CAS LC25 (24H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
CAS LC25 (48H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	250
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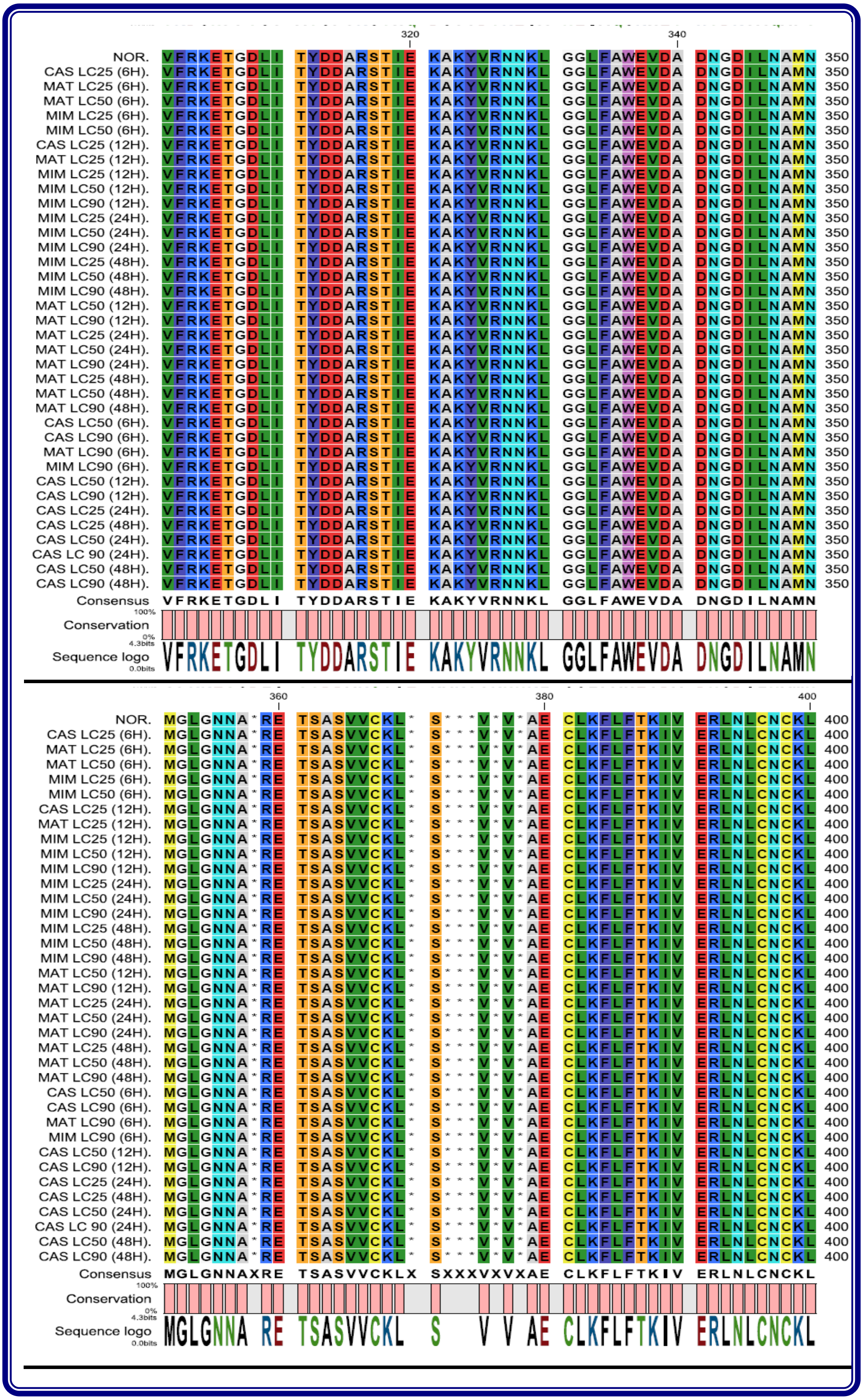
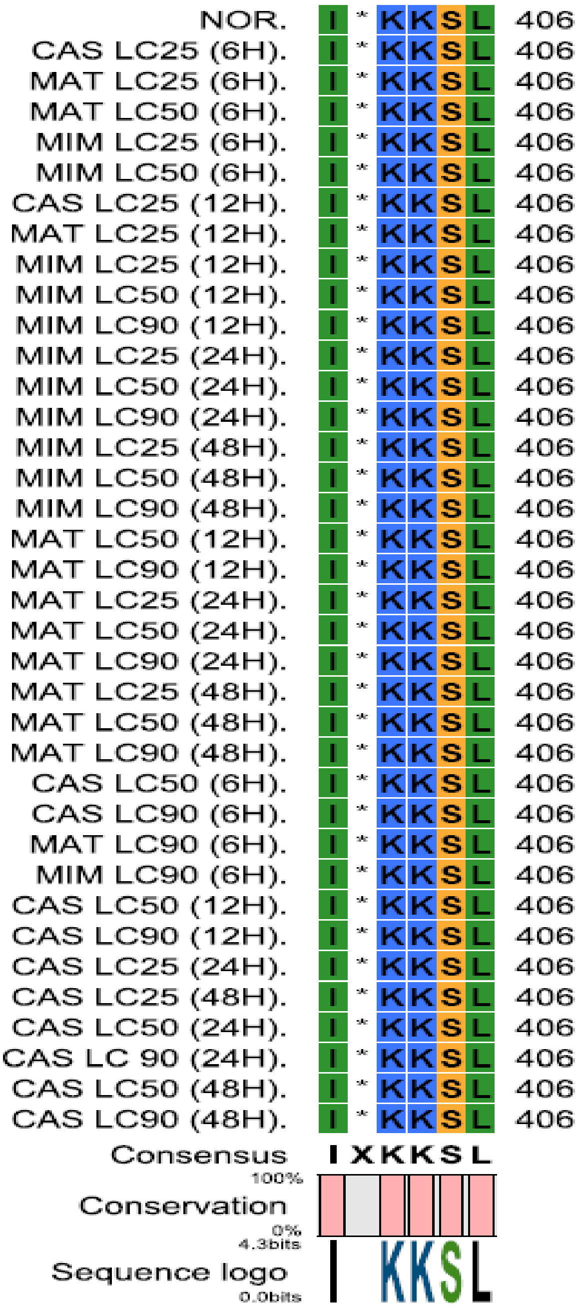
CAS LC 90 (24H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRRWTGVNG	250
CAS LC50 (48H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRRWTGVNG	250
CAS LC90 (48H).	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRRWTGVNG	250
Consensus	QAGLYAPAWN	PKETYTTDFG	VKFLLAQGVS	PKKIVVGVAM	YGRGWTGVNG	

	260		280		300	
NOR.	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC25 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC25 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC50 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC25 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC50 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC25 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC25 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC25 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC50 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC90 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC25 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC50 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC90 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC25 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC50 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC90 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC50 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC90 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC25 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC50 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC90 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC25 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC50 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC90 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC50 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC90 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MAT LC90 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
MIM LC90 (6H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC50 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC90 (12H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC25 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC25 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC50 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC 90 (24H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC50 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
CAS LC90 (48H).	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	300
Consensus	YKDGNPFTGV	ATGPVKGTWQ	DGVVDYREIA	NEIAQGKWEY	HYDKVAQAPY	



NOR.	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC25 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC25 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC50 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC25 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC50 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC25 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC25 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC25 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC50 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC90 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC25 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC50 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC90 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC25 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC50 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC90 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC50 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC90 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC25 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC50 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC90 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC25 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC50 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC90 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC50 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC90 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MAT LC90 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
MIM LC90 (6H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC50 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC90 (12H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC25 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC25 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC50 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC 90 (24H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC50 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
CAS LC90 (48H).	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	350
Consensus	VFRKETGDLI	TYDDARSTIE	KAKYVRNNKL	GGLFAWEVDA	DNGDILNAMN	

NOR.	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC25 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC25 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC50 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC25 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC50 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC25 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC25 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC25 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC50 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC90 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC25 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC50 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC90 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC25 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC50 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC90 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC50 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC90 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC25 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC50 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC90 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC25 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC50 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC90 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC50 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC90 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MAT LC90 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
MIM LC90 (6H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC50 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC90 (12H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC25 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC25 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC50 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC 90 (24H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC50 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
CAS LC90 (48H).	MGLGNNA*RE	TSASVVCKL*	S***V*V*AE	CLKFLFTKIV	ERLNLCNCKL	400
Consensus	MGLGNNAXRE	TSASVVCKLX	SXXXVXVXAE	CLKFLFTKIV	ERLNLCNCKL	



NOR.	I	*	K	K	S	L	406
CAS LC25 (6H).	I	*	K	K	S	L	406
MAT LC25 (6H).	I	*	K	K	S	L	406
MAT LC50 (6H).	I	*	K	K	S	L	406
MIM LC25 (6H).	I	*	K	K	S	L	406
MIM LC50 (6H).	I	*	K	K	S	L	406
CAS LC25 (12H).	I	*	K	K	S	L	406
MAT LC25 (12H).	I	*	K	K	S	L	406
MIM LC25 (12H).	I	*	K	K	S	L	406
MIM LC50 (12H).	I	*	K	K	S	L	406
MIM LC90 (12H).	I	*	K	K	S	L	406
MIM LC25 (24H).	I	*	K	K	S	L	406
MIM LC50 (24H).	I	*	K	K	S	L	406
MIM LC90 (24H).	I	*	K	K	S	L	406
MIM LC25 (48H).	I	*	K	K	S	L	406
MIM LC50 (48H).	I	*	K	K	S	L	406
MIM LC90 (48H).	I	*	K	K	S	L	406
MAT LC50 (12H).	I	*	K	K	S	L	406
MAT LC90 (12H).	I	*	K	K	S	L	406
MAT LC25 (24H).	I	*	K	K	S	L	406
MAT LC50 (24H).	I	*	K	K	S	L	406
MAT LC90 (24H).	I	*	K	K	S	L	406
MAT LC25 (48H).	I	*	K	K	S	L	406
MAT LC50 (48H).	I	*	K	K	S	L	406
MAT LC90 (48H).	I	*	K	K	S	L	406
CAS LC50 (6H).	I	*	K	K	S	L	406
CAS LC90 (6H).	I	*	K	K	S	L	406
MAT LC90 (6H).	I	*	K	K	S	L	406
MIM LC90 (6H).	I	*	K	K	S	L	406
CAS LC50 (12H).	I	*	K	K	S	L	406
CAS LC90 (12H).	I	*	K	K	S	L	406
CAS LC25 (24H).	I	*	K	K	S	L	406
CAS LC25 (48H).	I	*	K	K	S	L	406
CAS LC50 (24H).	I	*	K	K	S	L	406
CAS LC 90 (24H).	I	*	K	K	S	L	406
CAS LC50 (48H).	I	*	K	K	S	L	406
CAS LC90 (48H).	I	*	K	K	S	L	406
Consensus	I	X	K	K	S	L	

Fig. (63): Shows the alignment of the encoded amino acids of the vira-like chitinas gene sequences for the studied groups.

From all the above results we can recognized that we have seven groups of mutated vira-like chitinase gene resulted from the treatments of the 2nd larval instar with the different sub lethal doses of the used IGRs at the different times intervals post treatments in addition to the normal one **Table (40)**.

The 1st group (**G_1**) includes the treated larvae with the following doses, LC_{25} of Cascade, Match and Mimic after 6&12 hrs. respectively post treatments as well as LC_{50} of Match and Mimic after 6 hrs. The 2nd group (**G_2**) includes the treated larvae with the LC_{90} of Mimic after 6hrs. post treatment. The 3rd group (**G_3**) includes the treated larvae with the following doses, LC_{50} and LC_{90} of Cascade after 6&12 hrs. post treatments, LC_{90} of Match after 6hrs. post treatment and LC_{25} of Cascade after 24&48 hrs. post treatments. The 4th group (**G_4**) includes the treated larvae with the following doses, LC_{50} and LC_{90} of Match after 12hrs. post treatments as well as LC_{25} of Match after 24&48 hrs. post treatments. The 5th group (**G_5**) includes the treated larvae with the following doses, LC_{50} and LC_{90} of Mimic after 12, 24 and 48 hrs. post treatments and LC_{25} of Mimic after 24& 48 hrs. post treatments. The 6th group (**G_6**) includes the treated larvae with the following doses, LC_{50} and LC_{90} of Cascade after 24& 48 hrs. post treatments. Finally the 7th group (**G_7**) that includes the treated larvae with the following doses, LC_{50} and LC_{90} of Match after 24& 48hrs. post treatments.

	DNA sequence				
Treated groups	**Nucleotide change**	**Nucleotide change position**	**Amino acid change**	**Amino acid position**	**Restriction enzymes**
G1	———	———	———	———	*EcoRI* *BbsI*
G2	T ► C	194	I ► T	65	*EcoRI* *BbsI*
G3	T ► C G ► C	194 468	I ► T L ► F	65 156	*EcoRI* *BbsI*
G4	G ► C T ► C	468 701	L ► F I ► T	156 234	*EcoRI* *BbsI*
G5	T ► C G ► C	384 468	L ► F	156	*EcoRI* *BbsI*
G6	T ► C G ► C G ► C	194 468 730	I ► T L ► F G ► R	65 156 244	*EcoRI* *BbsI*
G7	G ► C G ► C T ► C	260 468 701	R ► T L ► F I ► T	87 156 234	*EcoRI* *BbsI*

Table (39): groups of mutated vira-like chitinase gene resulted from the treatments of the 2nd larval instar with the different sub lethal doses of the used IGRs at the different times intervals post treatments.

4.5.3 The expected protein charges:-

The protein charge of the seven studied groups was evaluated and plotted against the pH compared with normal by using CLC Main Workbench program (version 5.5).

It is clearly evident that the protein charges of the 1st group and the 2nd group were with the same charges as well as normal at different pH values, **Fig (64)**. The protein charge of the 4th group has slightly low charge 56.340 as compared with normal 57.192 at pH2, Fig. **(65)**.

Fig. (66) Showed that the 3rd group has the highest charges as compared with normal at the different PH values. The remaining groups (5th , 6th and 7th) showed low charges as compared with normal respectively **Fig. (67), (68)** and **Fig. (69)** respectively. **Fig. (70)** shows the protein charges of all groups collected with each others.

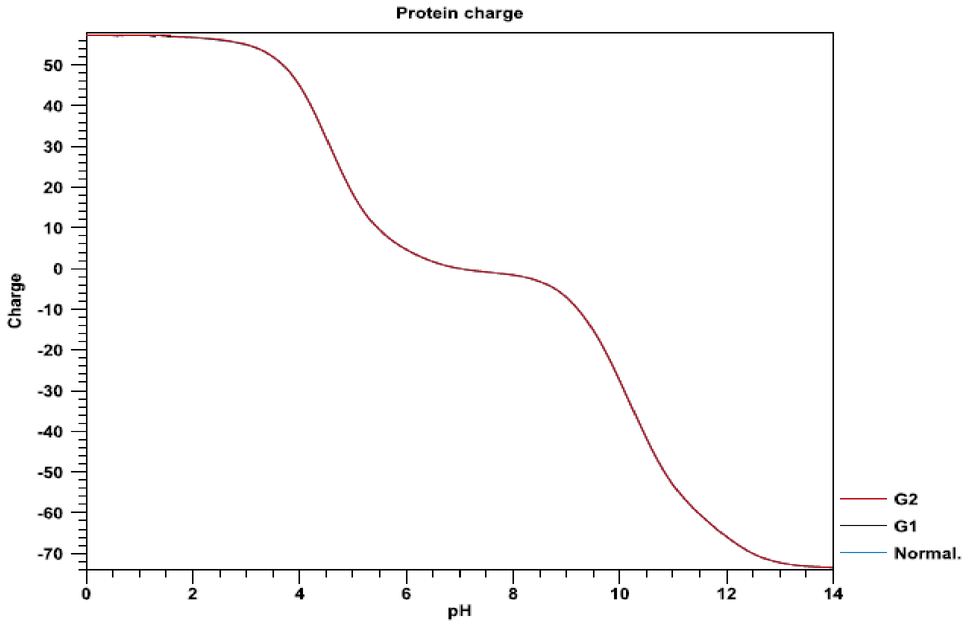


Fig. (64) : protein charge of 1st group and 2nd group.

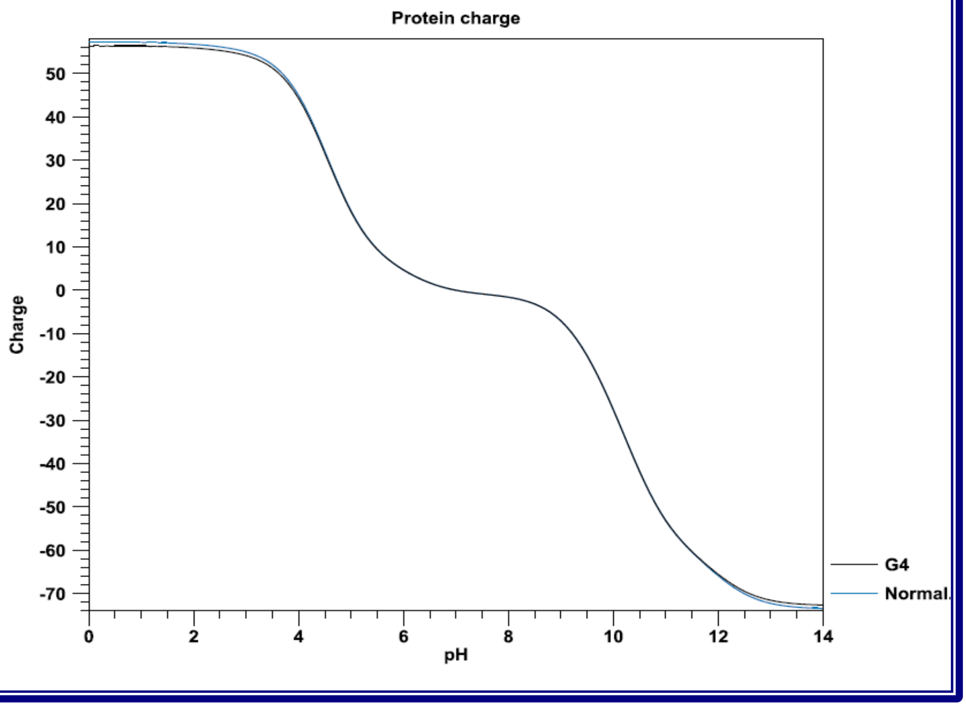


Fig. (65) : protein charge of 4th group.

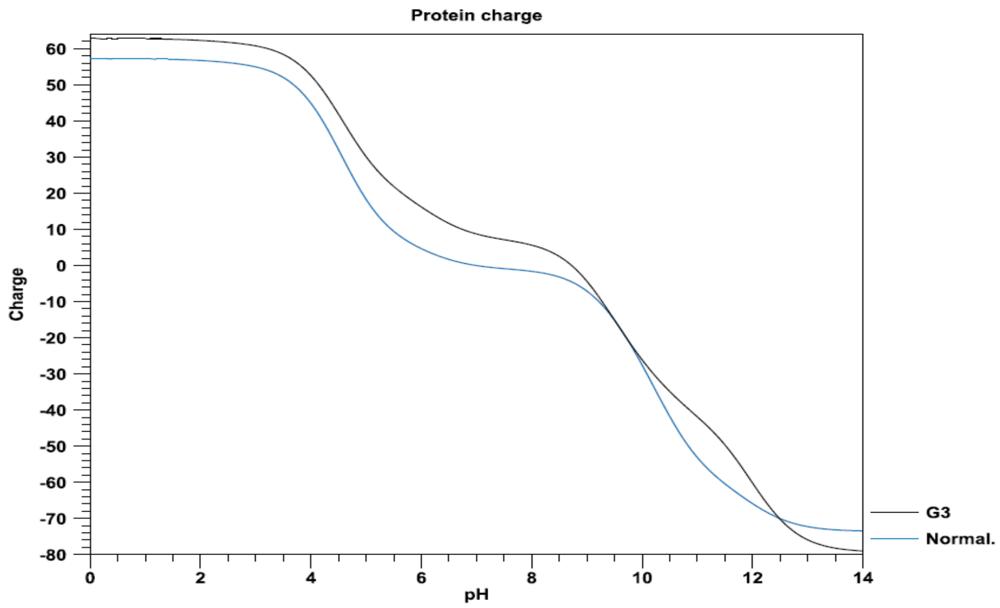


Fig. (66) : protein charge of 3rd group.

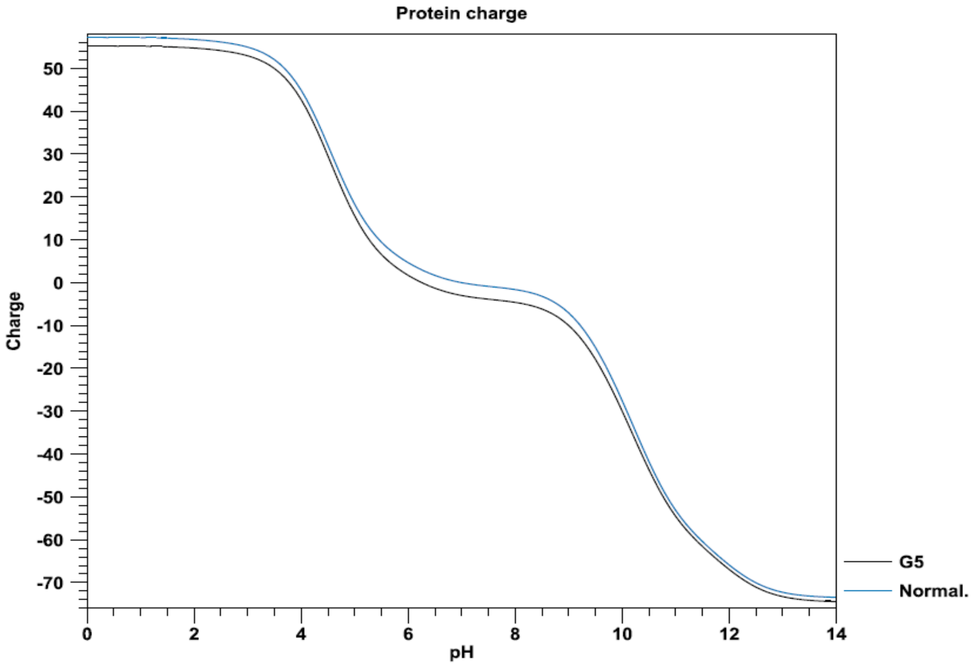


Fig. (67) : protein charge of 5th group.

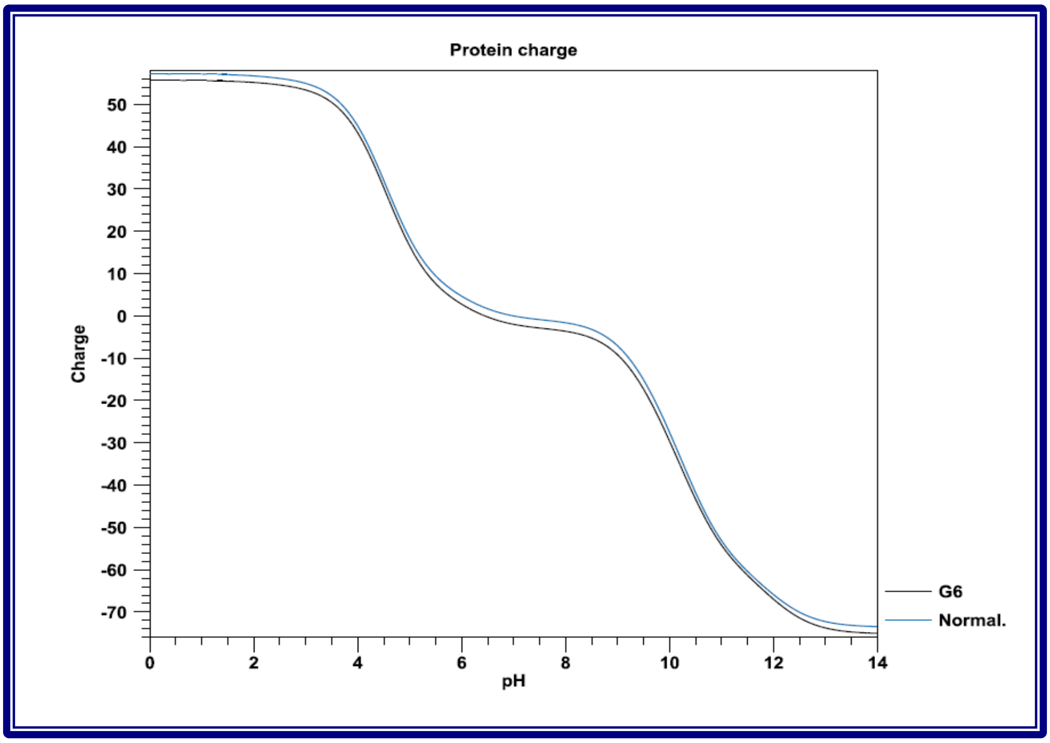


Fig. (68) : protein charge of 6th group.

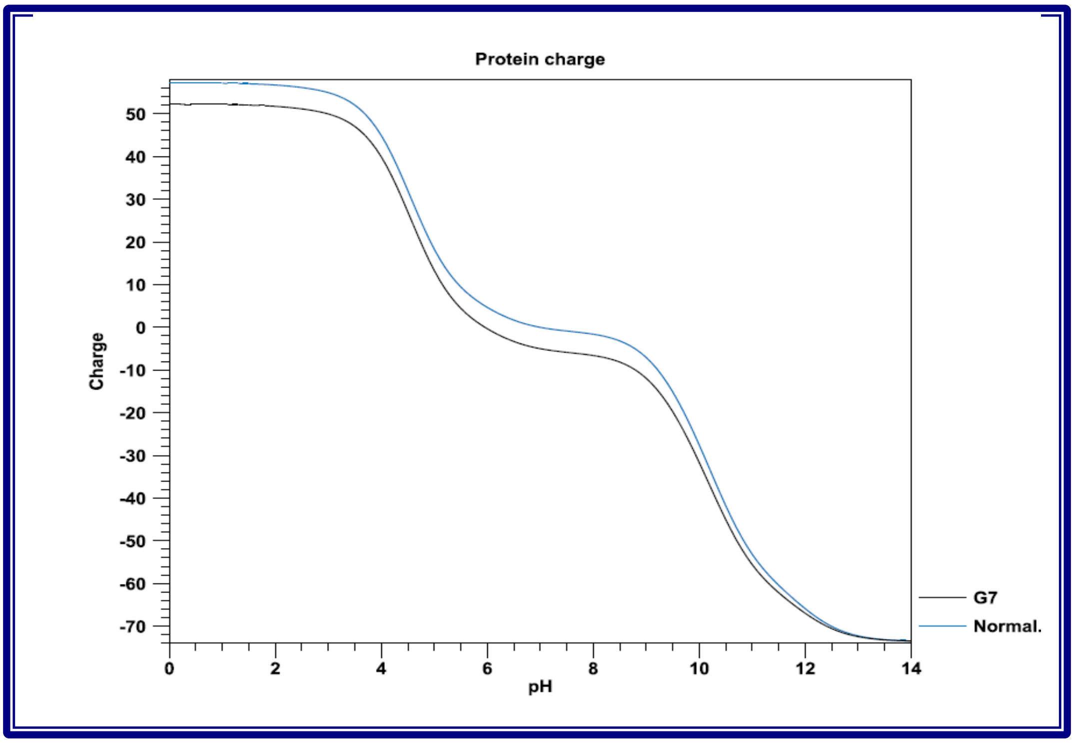


Fig. (69) : protein charge of 7th group.

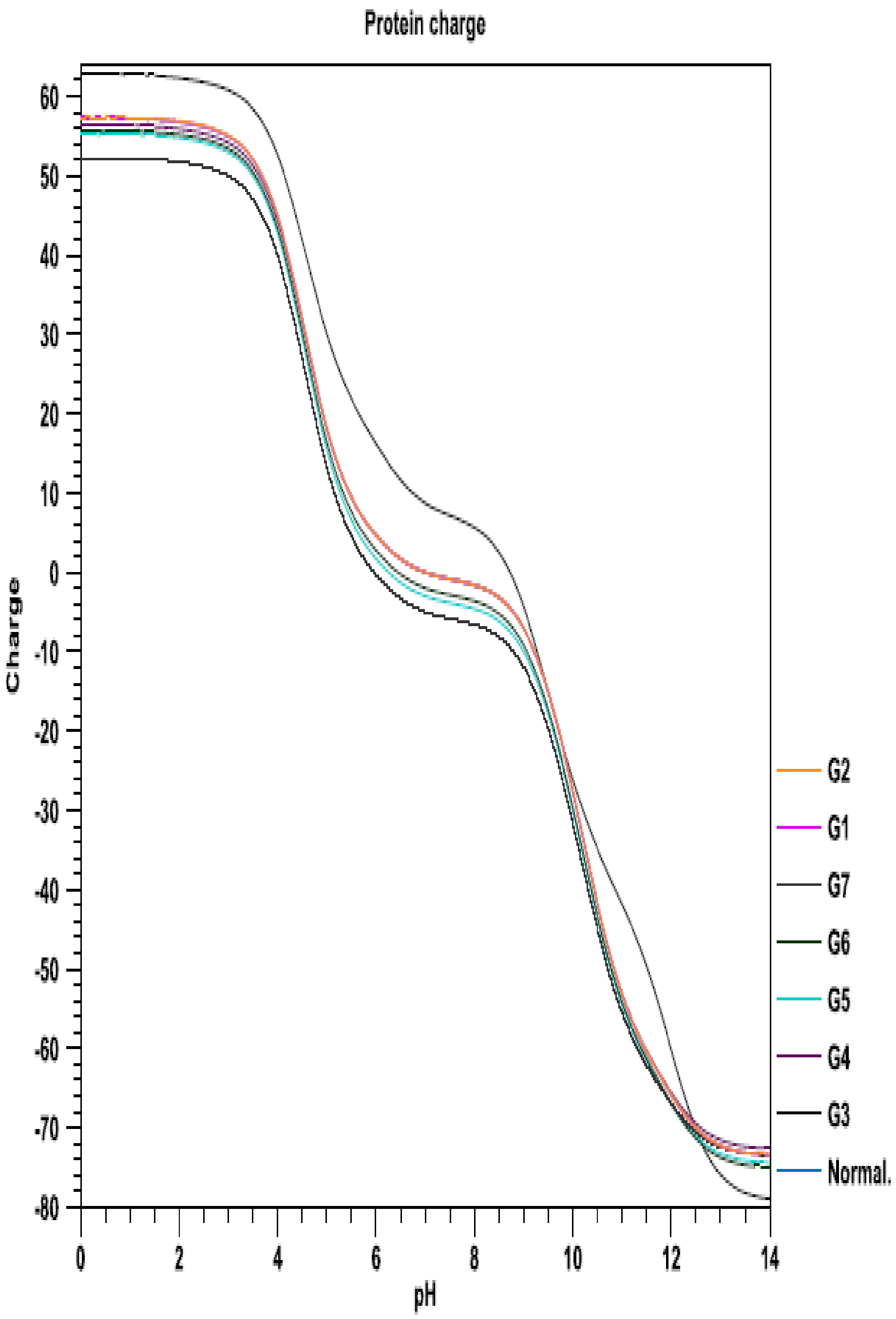


Fig. (70) : protein charges of all groups.

4.5.4 RNA secondary structure prediction:-

RNA which transcribed from vira-like chitinase gene is a single strand, this strand has several complementary sequences (paired) stems and single stranded (unpaired) loops. The stems with complementary nucleotides and resulted loops formed a distinct structure known as RNA secondary structure. As the yielding protein of the mutated vira-like chitinase gene has been changed, the RNA secondary structure also is expected to be changed.

The number of RNA secondary structure stems is varied in the different studied groups and most of them differed in their nucleotide sequences, positions and free energy **Table (40)**.

For each studied group, the RNA secondary structure stems nucleotide sequences, positions and free energy were recorded as well as the distinct secondary structure was drawn using a computer based online program known as gene bee RNA secondary structure prediction (Genebee service, http://www.genebee.msu.ru/cgi-bin/nph-malign.pl) **Figures,(71-72-73-74-75-76-77).**

Table (40): Shows the sequence, position and free energy of the different stems of the predicted RNA structure of the fetuses of the different studied groups.

Free energy Kkal/mol	Stem sequence	position	STEM NUMBER							
			N	G1	G2	G3	G4	G5	G6	G7
-16.700000	CCTGGTGG GGACCACC	340-347 604-597						1		1
-16.400000	TCCTTGGG AGGAACCC	81-88 638-631						2		
-16.200000	CAGTCTCC GTCAGAGG	18-25 919-912						3		
-15.500000	CTGCCAAG GACGGTTC	440-447 810-803	1	1	1		1		1	
-15.500000	CTGCCAAG GACGGTTC	444-451 814-807				1				
-15.400000	ATCTTCTTGA TAGAAGAACT	544-553 701-692							2	
-14.800000	TGGACCA ACCTGGT	536-542 792-786	2	2	2		2		3	
-14.800000	TGGACCA ACCTGGT	540-546 796-790				2				
-14.200000	GAAGGCA CTTCCGT	13-19 203-197	3	3	3		3		4	2
-14.200000	GAAGGCA CTTCCGT	16-22 207-201				3				
-14.000000	GAGCCA CTCGGT	353-358 889-884	4	4	4		4		5	
-14.00000	GAGCCA CTCGGT	357-362 893-888				4				
-13.800000	TCTTCAGA AGAAGTCT	30-37 193-186	5	5	5		5			3
-13.700000	TCTTCAGA AGAAGTCT	30-37 193-186							6	
-13.700000	TCTTCAGA AGAAGTCT	33-40 197-190				5				
-13.200000	GAGACC CTCTGG	920-925 1059-1054	6	6	6		6		7	4
-13.200000	GAGACC CTCTGG	924-925 1063-1085				6				
-13.100000	ATCTTCTTG TAGAAGAAC	544-552 701-693	7	7	7					
-13.100000	ATCTTCTTG TAGAAGAAC	548-556 705-697				7				
-13.000000	TGGACC ACCTGG	214-219 792-787								5
-12.400000	AGCTGAT TCGACTA	401-407 484-478						4		6
-12.300000	TCTTCTTG AGAAGAAC	233-240 700-693								7
-12.300000	TCTTCTTG AGAAGAAC	545-552 700-693					7			
-12.200000	CCCTGA GGGACT	375-380 857-852	8	8	8		8		8	
-12.200000	CCCTGA GGGACT	379-384 861-856				8				
-12.100000	GAGACA CTCTGT	258-263 1146-1141	9	9	9		9		9	
-12.100000	GAGACA CTCTGT	262-267 1150-1145				9				

Continued table (40)

Free energy Kkal/mol	Stem sequence	position	STEM NUMBER							
			N	G1	G2	G3	G4	G5	G6	G7
-12.000000	CTCTTCA GAGAAGT	92-98 260-254						5		
-12.000000	CCAACC GGTTGG	356-361 492-487						6		8
-11.700000	GCTGG CGACC	604-608 623-619	10	10	10		10			
-11.700000	GCTGG CGACC	608-612 627-623				10				
-11.600000	GCTACA CGATGT	749-754 782-777	11	11	11		11	7	10	10
-11.600000	GCTACA CGATGT	753-758 786-781				11				
-11.600000	TTGGCA AACCGT	804-809 842-837						8		9
-11.500000	GGCTG CCGAC	603-607 691-687							11	
-11.300000	ACTGGA TGACCT	736-741 794-789						9		
-11.200000	AAGCTC TTCGAG	521-526 577-572						10		11
-11.100000	CTTCCT GAAGGA	288-293 912-907	12	12	12		12		12	
-11.100000	CTTCCT GAAGGA	292-297 916-911				12				
-11.100000	GCCC CGGG	689-692 858-855						11		
-11.100000	GCAAGA CGTTCT	175-180 199-194						12		
10.800000	TCTTGC AGAACG	40-45 180-175	13	13	13		13		13	12
-10.800000	TCTTGC AGAACG	43-48 184-179				13				
-10.700000	AGGGT TCCCA	140-144 221-217						13		
-10.600000	TTCCTG AAGGAC	667-672 911-906						14		
-10.600000	TCAAGG AGTTCC	281-286 296-291						15		13
-10.300000	GACAC CTGTG	586-590 663-659	14	14	14		14			
-10.300000	GAGAC CTCTG	920-924 1146-1142						16		
-10.300000	GACAC CTGTG	590-594 667-663				14				
-10.300000	GAGAC CTCTG	913-917 1146-1142								14
-10.200000	GACAC CTGTG	258-262 687-683								15
-10.000000	TGAAGA ACTTCT	54-59 98-93					15		14	16
-10.000000	AGATGT TCTACA	442-427 474-469						17		17
-9.900000	GAACAA CTTGTT	978-983 1002-997	15	15	15		16	18	15	19

Continued table (40)

Free energy Kkal/mol	Stem sequence	position	STEM NUMBER							
			N	**G1**	**G2**	**G3**	**G4**	**G5**	**G6**	**G7**
-9.900000	GAACAA CTTGTT	982-987 1006-1001				15				
-9.900000	TTGACA AACTGT	320-325 665-660								18
-9.800000	CTGG GACC	605-608 622-619						19		21
-9.800000	TTCACT AAGTGA	241-246 257-252							16	20
-9.700000	AAACCT TTTGGA	100-105 116-111	16	16	16		17	20	17	23
-9.700000	AAACCT TTTGGA	104-109 120-115				16				
-9.700000	CGTTG GCAAC	702-706 748-744					18			22
-9.500000	GGCT CCGA	87-90 134-131	17	17	17					
-9.500000	GGCT CCGA	91-94 138-135				17				
-9.500000	CAGTC GTCAG	278-282 919-915							18	
-9.500000	TTGGG AACCC	330-334 635-631								24
-9.400000	ACACC TGTGG	261-265 320-316						21		
-9.300000	TTCACT AAGTGA	241-246 257-252	18	18	18					
-9.300000	CTGCT GACGA	473-477 531-527	19	19	19	19	20		19	
-9.300000	TTCACT AAGTGA	245-250 261-256				18	19			
-9.200000	AGCC TCGG	131-134 225-222						22		
-9.000000	CGCG GCGC	942-945 1020-1017	20	20	20		21	23	20	25
-9.000000	CGCG GCGC	946-949 1024-1021				20				
-8.800000	GAACG CTTGC	126-130 234-230						24		
-8.700000	CAAAGT GTTTCA	63-68 658-653						25		
-8.700000	GGTG CCAC	682-685 896-893						26		
-8.700000	GGTG CCTC	882-885 896-893								26
-8.600000	CGTTC GCAAG	230-234 1175-1171	21	21	21		22		21	
-8.600000	CCTTG GGAAC	86-90 146-142				21				
-8.600000	CCTTG GGAAC	82-86 142-138	22	22	22					
-8.600000	CTTGG GAACC	596-600 640-636				22				
-8.600000	CTTGG GAACC	592-596 636-632	23	23	23		23			

Continued table (40)

Free energy Kkal/mol	Stem sequence	position	STEM NUMBER							
			N	G1	G2	G3	G4	G5	G6	G7
-8.300000	GGTG CCAC	343-346 896-893	24	24	24		24		22	
-8.300000	GGTG CCAC	347-350 900-897				23				
-8.300000	CAAGA GTTCT	495-499 595-591						27		27
-8.300000	GGC CCG	49-51 154-152	25	25	25					
-8.300000	GGC CCG	52-54 158-156				24				
-8.200000	GCAAT CGTTA	715-719 851-847						28		
-8.100000	CTTCA GAAGT	275-279 1076-1072	26	26	26		25			
-8.100000	CTTCA GAAGT	279-283 1080-1076				25				
-8.100000	CGTTG GCAAC	702-706 748-744	27	27	27				23	
-8.100000	CGTTG GCAAC	706-710 752-748				26				
-8.000000	TCATC AGTAG	118-122 163-159					26		24	28
-8.000000	CTTGA GAACT	549-553 570-566						29		29
-8.000000	AGGA TCCT	635-638 671-668							25	
-7.900000	TCTAC AGATG	392-396 824-820	28	28	28		27		26	
-7.900000	TCTAC AGATG	396-400 828-824				27				
-7.700000	ATTACG TAATGC	928-933 1117-1112						30		
-7.700000	GTCA CAGT	117-120 249-246						31		
-7.600000	GGAT CCTA	215-218 234-231				28				
-7.600000	CACA GTGT	677-680 903-900						32		
-7.600000	TGTTG ACAAC	204-208 1164-1160								30
-7.500000	CCAA GGTT	153-156 209-206						33		
-7.400000	AGCT TCGA	401-404 437-434	29	29	29		28		27	
-7.400000	AGCT TCGA	405-408 441-438				29				
7.400000	GACG CTGC	382-385 399-396						34		
7.400000	AGCT TCGA	481-484 525-522	30	30	30		29		28	
-7.400000	AAGTTA TTCAAT	68-73 153-148				30				
-7.400000	AGCT TCGA	485-488 529-526				31				

Continued table (40)

Free energy Kkal/mol	Stem sequence	position	STEM NUMBER							
			N	G1	G2	G3	G4	G5	G6	G7
-7.300000	GTTC CAAG	235-238 1178-1175				32				
-7.300000	GCC CGG	132-134 153-151					30		29	31
-7.300000	GAGT CTCA	555-558 592-589							30	
-7.200000	TGAC ACTG	561-564 688-685	31	31	31		31			
-7.200000	TGAC ACTG	565-568 692-689				33				
-7.200000	CATATT GTATAA	1032-1037 1053-1048						35		
-7.200000	TGAC ACTG	651-654 664-661							31	
-7.200000	GAAC CTTG	800-803 906-903								32
-7.100000	TATCA ATAGT	868-872 880-876	32	32	32		32		32	34
-7.100000	GGA CCT	54-56 76-74			33					
-7.100000	TATCA ATAGT	872-876 884-880				34				
-7.100000	TTGC AACG	848-851 860-857								33
-7.000000	AGATT TCTAA	8-12 1184-1180	33	33	34			36	33	35
-7.000000	GCT CGA	574-576 677-675	34	34	35		33			
-7.000000	GCT CGA	578-580 681-679				35				
-7.000000	GCT CGA	28-30 48-46						37		
-6.900000	CCC GGG	360-362 865-863	35	35	36		34		34	
-6.900000	CCC GGG	364-366 869-867				36				
-6.900000	ACAAT TGTTA	1022-1026 1108-1104						38		
-6.800000	GAAG CTTC	55-58 150-147	36	36						
-6.800000	CTTC GAAG	275-278 306-303						39		36
-6.800000	ACC TGG	263-265 318-316								37
-6.700000	CGC GCG	265-267 1089-1987	37	37	37		35			
-6.700000	CGC GCG	269-271 1093-1091				37				
-6.700000	AAACT TTTGA	1079-1083 1126-1122							35	
-6.600000	CCA GGT	443-445 452-450						40		38
-6.500000	AATTTTT TTAAAAA	1148-1154 1167-1161	38	38	38		37	41		

Continued table (40)

Free energy **Kkal/mol**	**Stem sequence**	**position**	**STEM NUMBER**							
			N	**G1**	**G2**	**G3**	**G4**	**G5**	**G6**	**G7**
-6.500000	AATTTTT TTAAAAA	1152-1158 1171-1165				38				
-6.500000	AGATT TCTAA	8-12 1184-1180					36			
-6.300000	GAC CTG	58-60 663-661						42		
-6.300000	CTTC GAAG	267-270 1076-1073							36	
-6.200000	AGAC TCTG	57-60 72-69			39					
-6.200000	CGA GCT	1088-1090 1113-1111							37	
-6.200000	TCC AGG	616-618 628-626							38	
-6.100000	GAT CTA	212-214 229-227	39	39	40		38		39	
-6.100000	GTC CAG	284-286 921-919				39				
-6.100000	GTC CAG	280-282 917-915	40	40	41		39			
-6.100000	ACC TGG	629-631 684-682							40	
TOTAL NUMBER OF STEMS										
			40	**40**	**41**	**39**	**39**	**42**	**40**	**38**

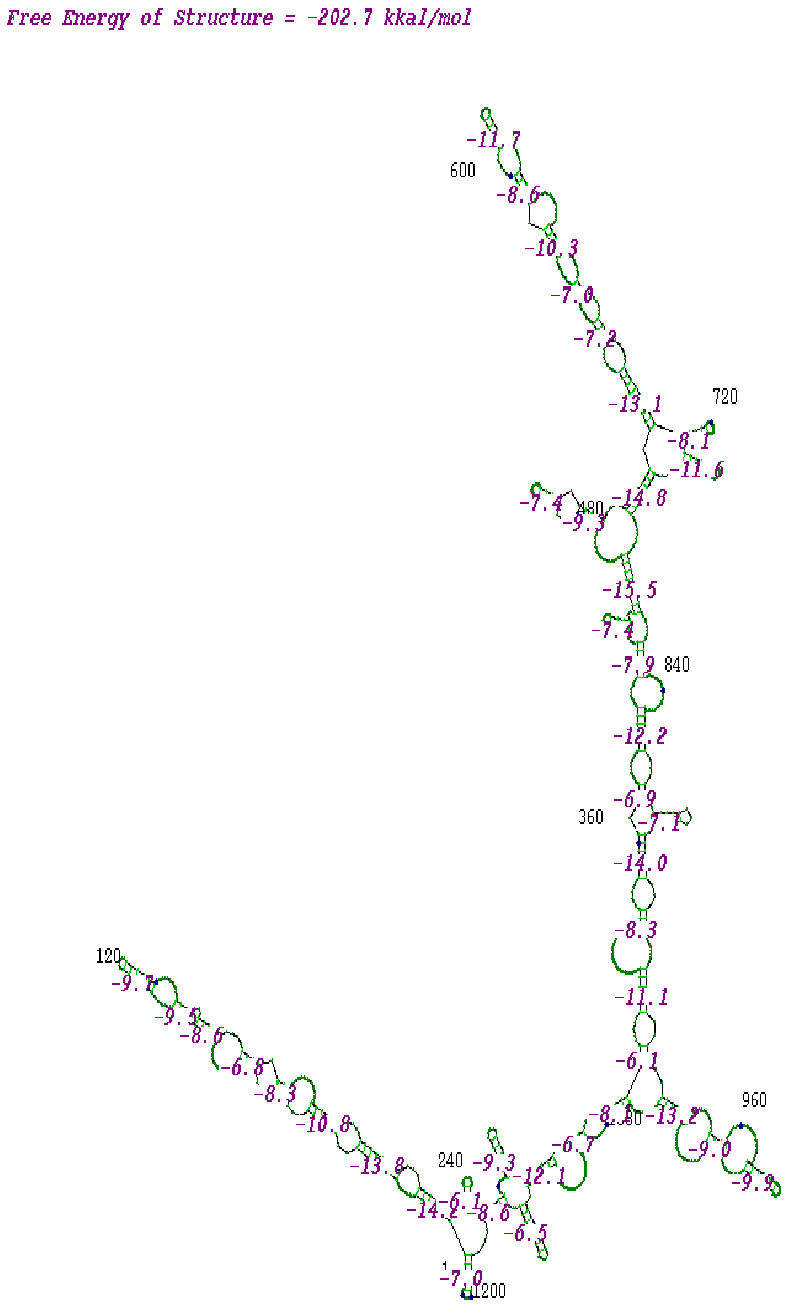


Fig. (71): Shows the structure of the predicted RNA structure of the normal and 1st group.

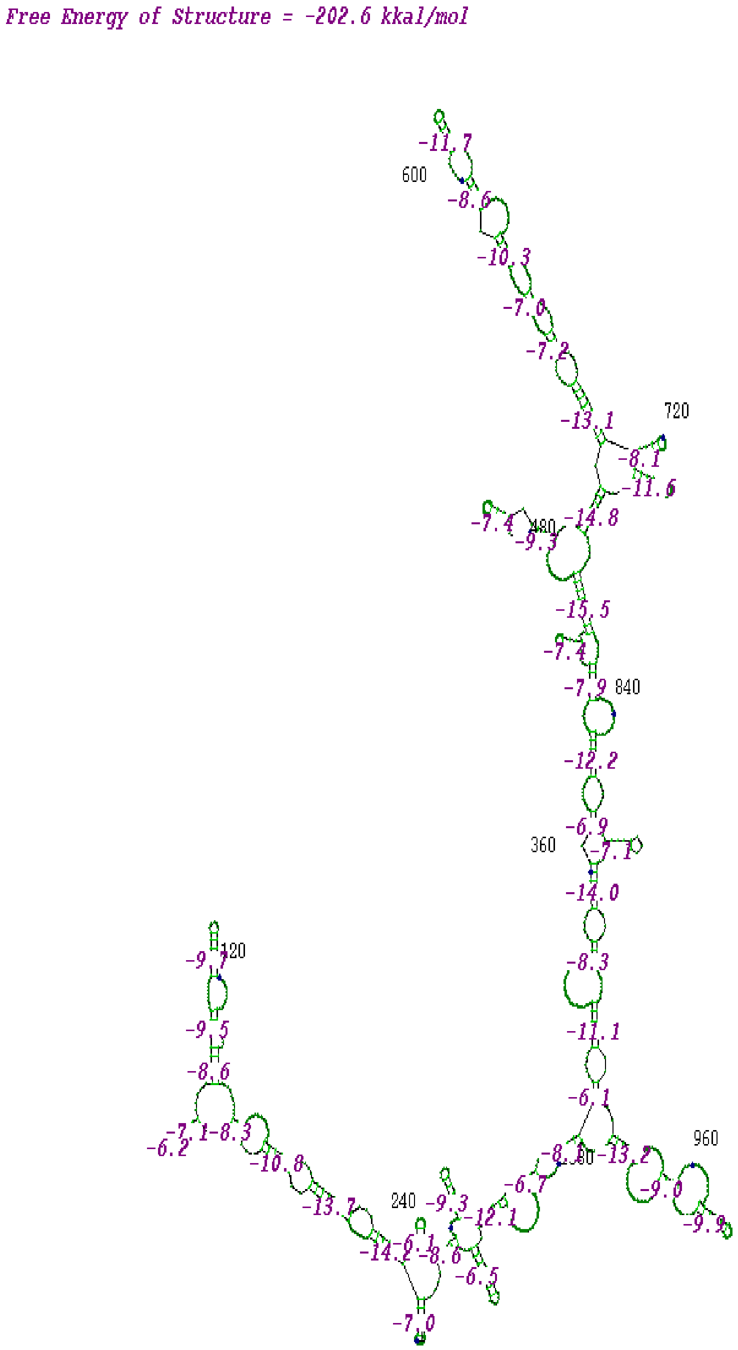


Fig. (72): Shows the structure of the predicted RNA structure of 2nd group.

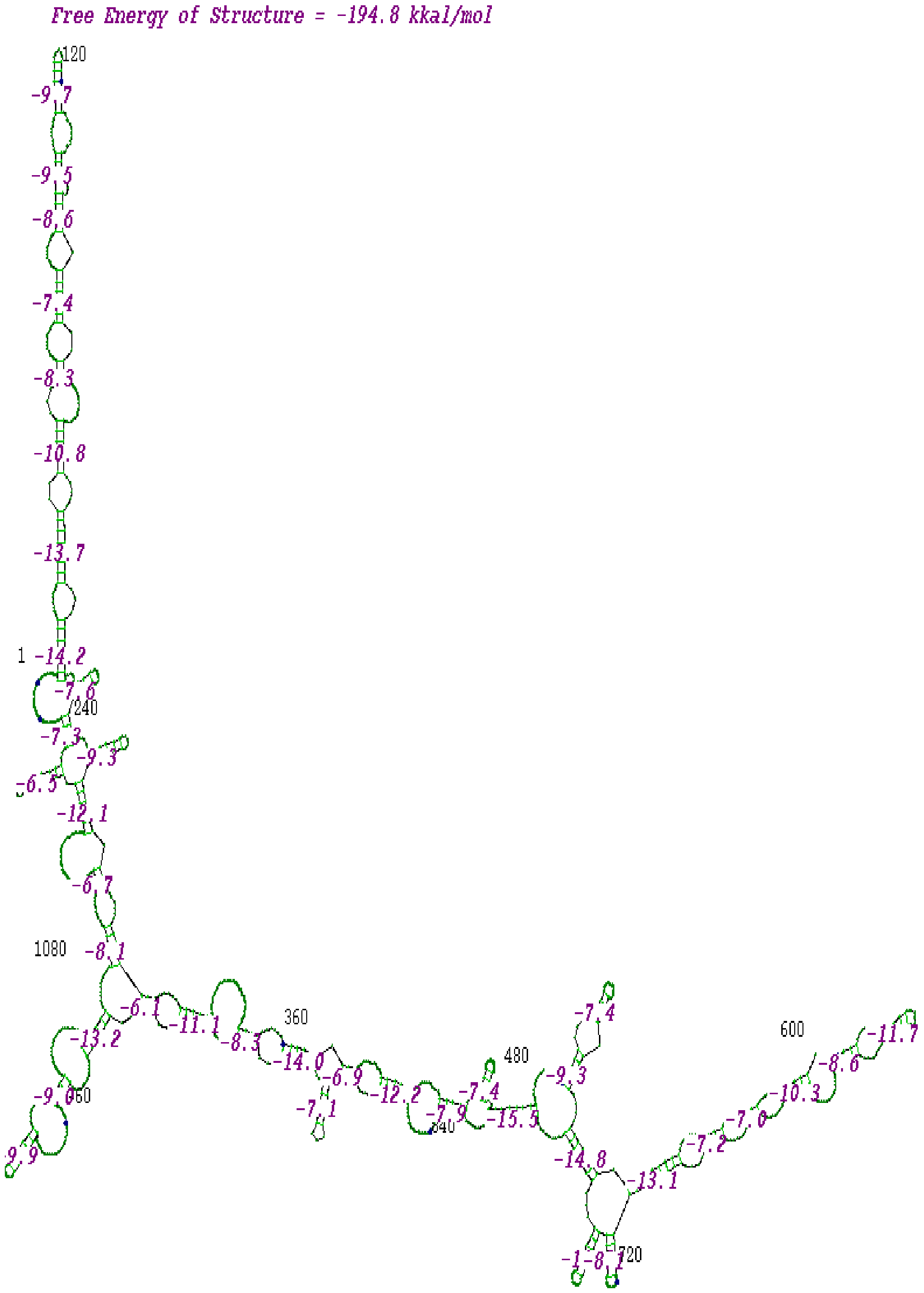


Fig. (73): Shows the structure of the predicted RNA structure of 3rd group.

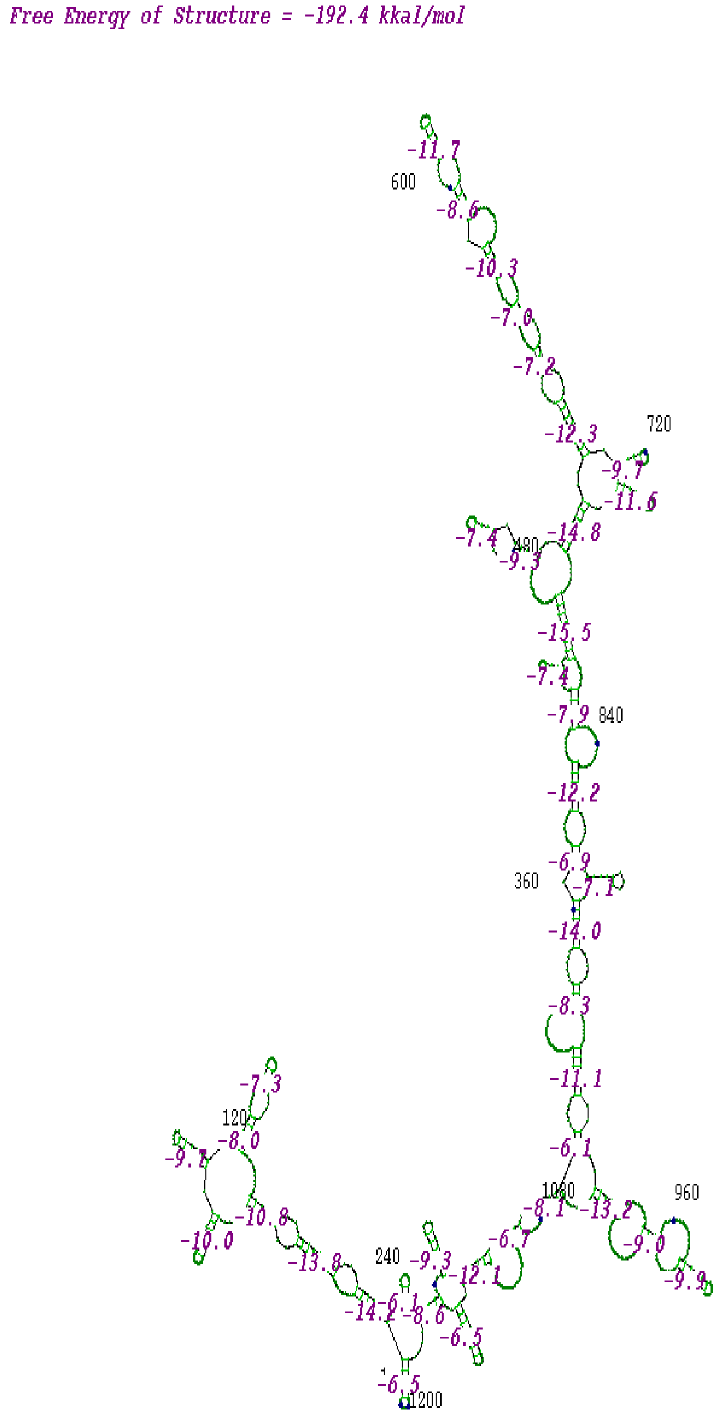


Fig. (74): Shows the structure of the predicted RNA structure of 4th group.

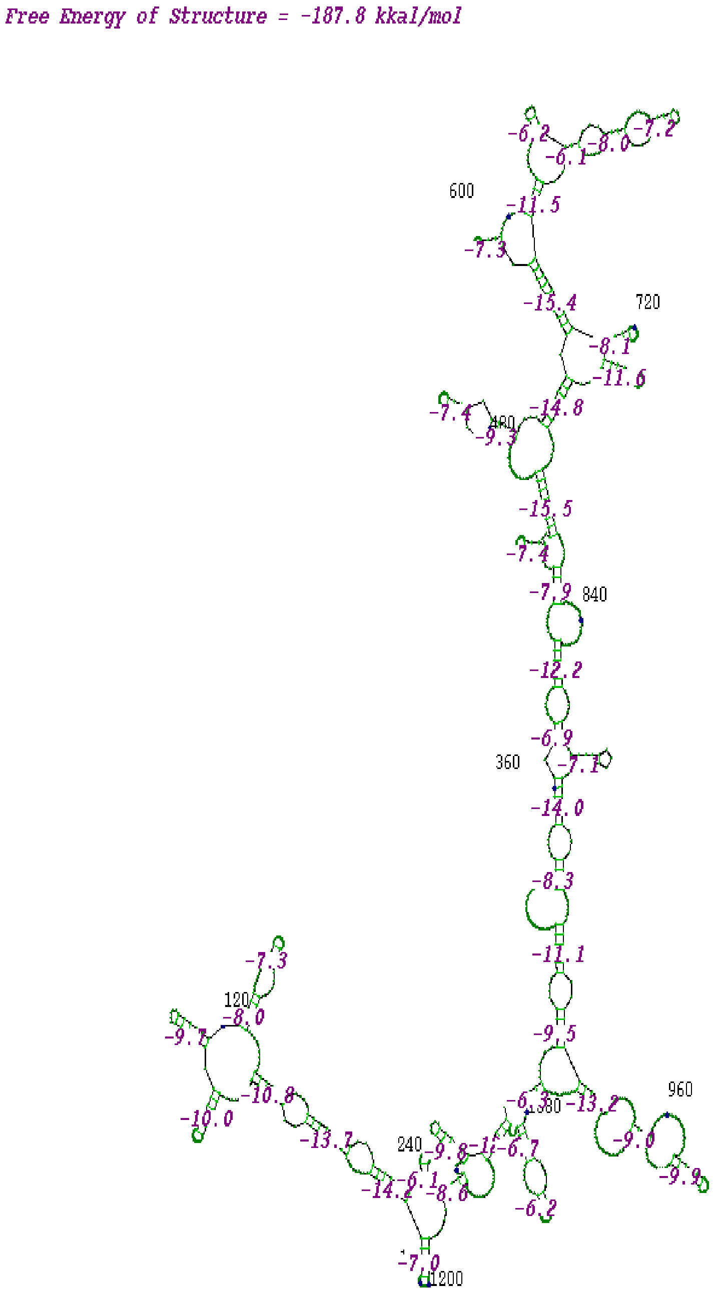


Fig. (75): Shows the structure of the predicted RNA structure of 5th group.

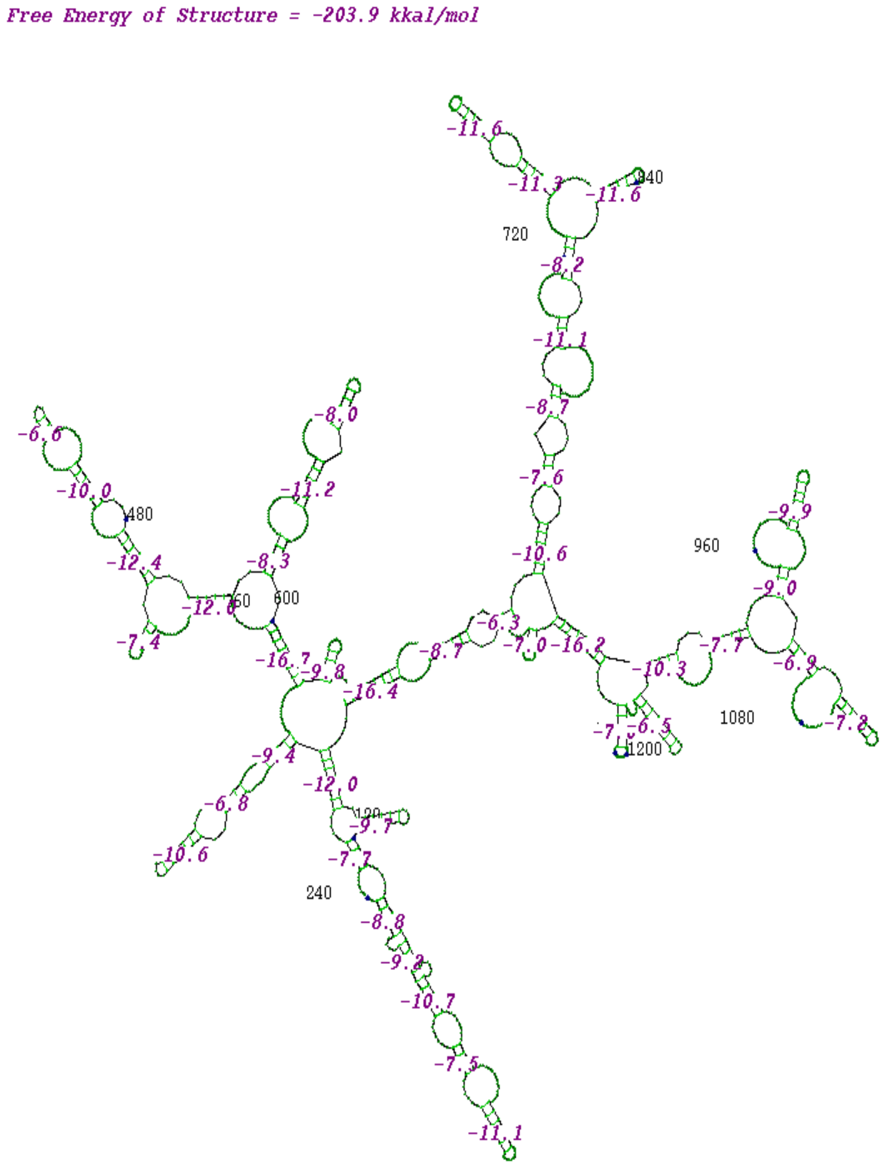


Fig. (76): Shows the structure of the predicted RNA structure of 6th group.

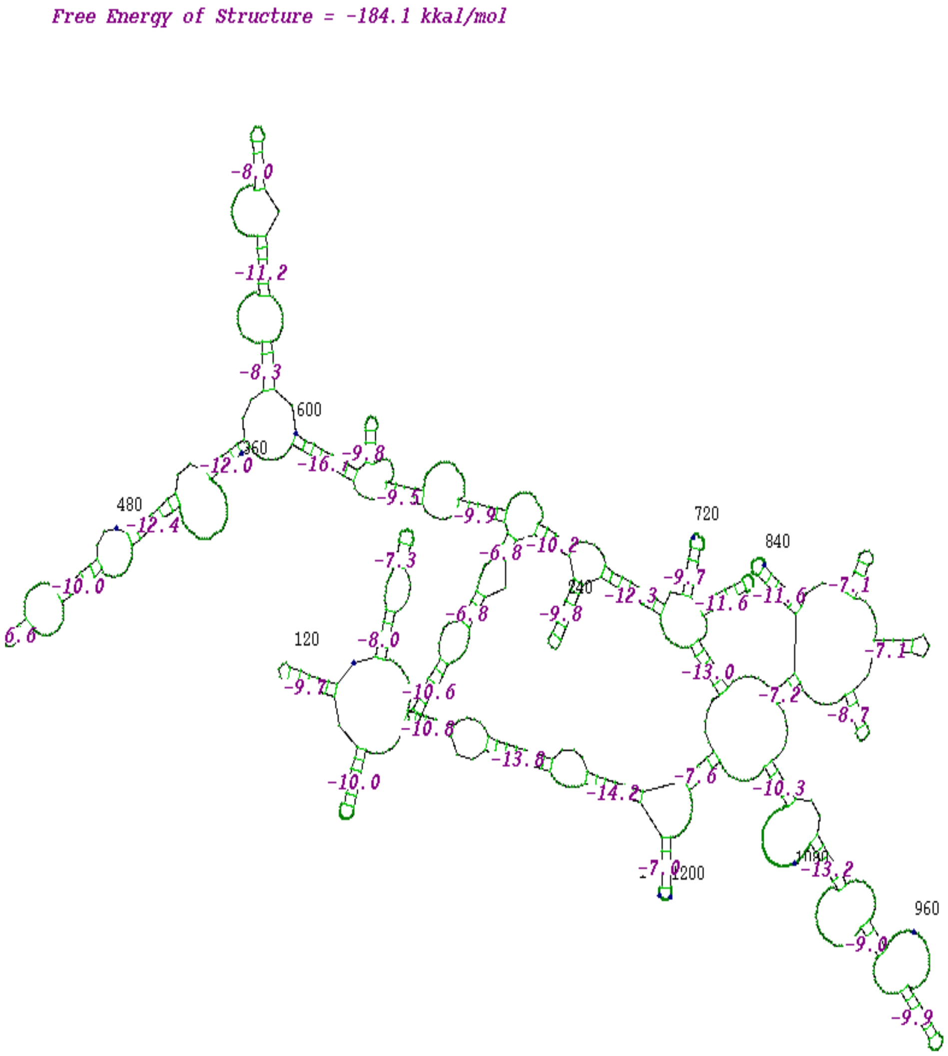


Fig. (77): Shows the structure of the predicted RNA structure of 7th group.

4.5.5 Phylogenetic tree prediction:-

Several different phylogenetic analyses were used for the sequence data of nuclear vira-like chitinase gene by using GeneBee Service (http://www.genebee.msu.ru/cgi-bin/nph-malign.pl).

Table (41) shows the distance matrix generated by genebee topological algorithm for vira-like chitinase gene of the seven studied groups, **Figures (66-67-68-69-70-71-72)** are represent the different shapes of the phylogenetic tree for the studied groups as compared with normal one dissending order on the bases of which group is nerest and mor similar to the normal one and so on.

Table (41): Distance matrix generated by genebee topological algorithm for vira-like chitinase gene of the seven studied groups.

	normal	G1	G2	G3	G4	G5	G6	G7
normal	0.000	0.999	0.999	0.999	0.999	0.999	0.999	0.999
G1	0.999	0.000	0.999	0.999	0.999	0.999	0.999	0.999
G2	0.999	0.999	0.000	0.998	0.999	0.999	0.998	0.999
G3	0.999	0.999	0.998	0.000	0.998	0.998	0.998	0.998
G4	0.999	0.999	0.999	0.998	0.000	0.998	0.998	0.998
G5	0.999	0.999	0.999	0.998	0.998	0.000	0.998	0.998
G6	0.999	0.999	0.998	0.998	0.998	0.998	0.000	0.998
G7	0.999	0.999	0.999	0.998	0.998	0.998	0.998	0.000

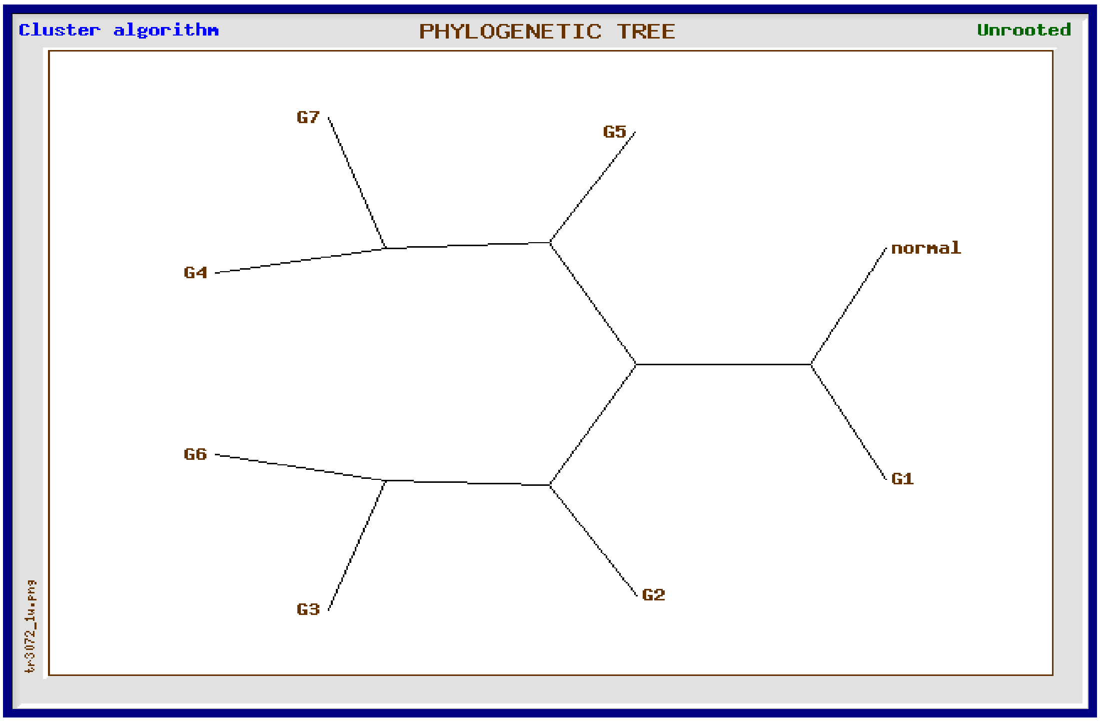


Fig. (78): A cladogram based on alignment of gene sequences of the seven studied groups; drawn by GeneBee program (Unrooted cluster algorithm).

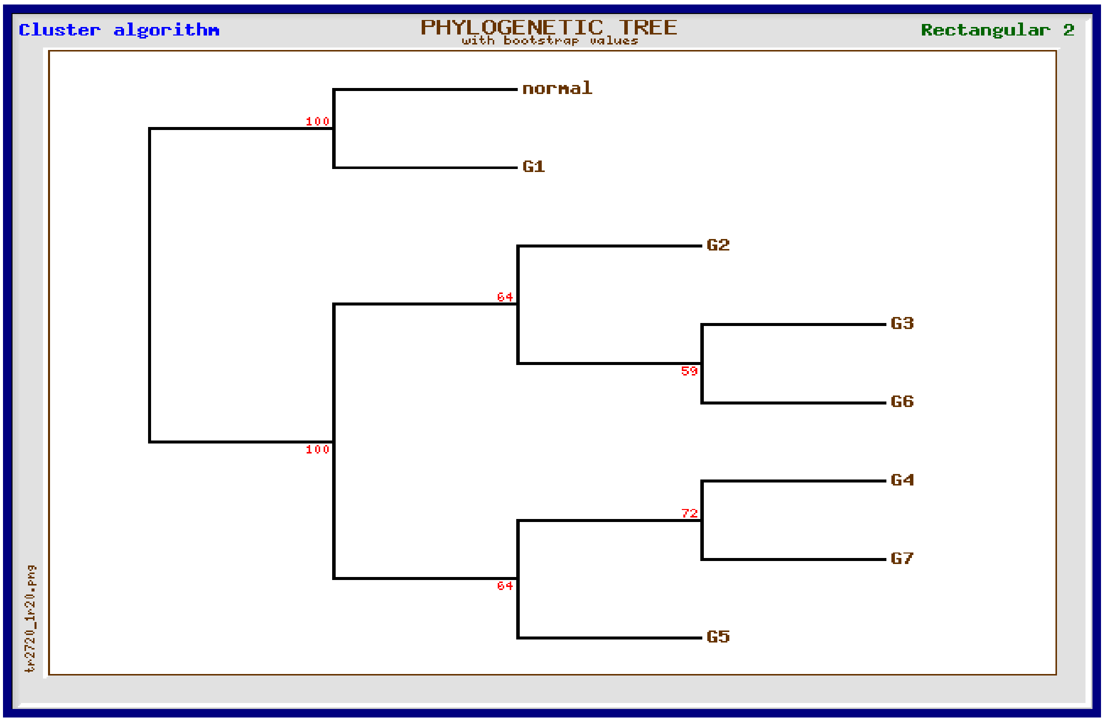


Fig. (79): A cladogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the seven studied groups; drawn by GeneBee program (Rectangular cluster algorithm).

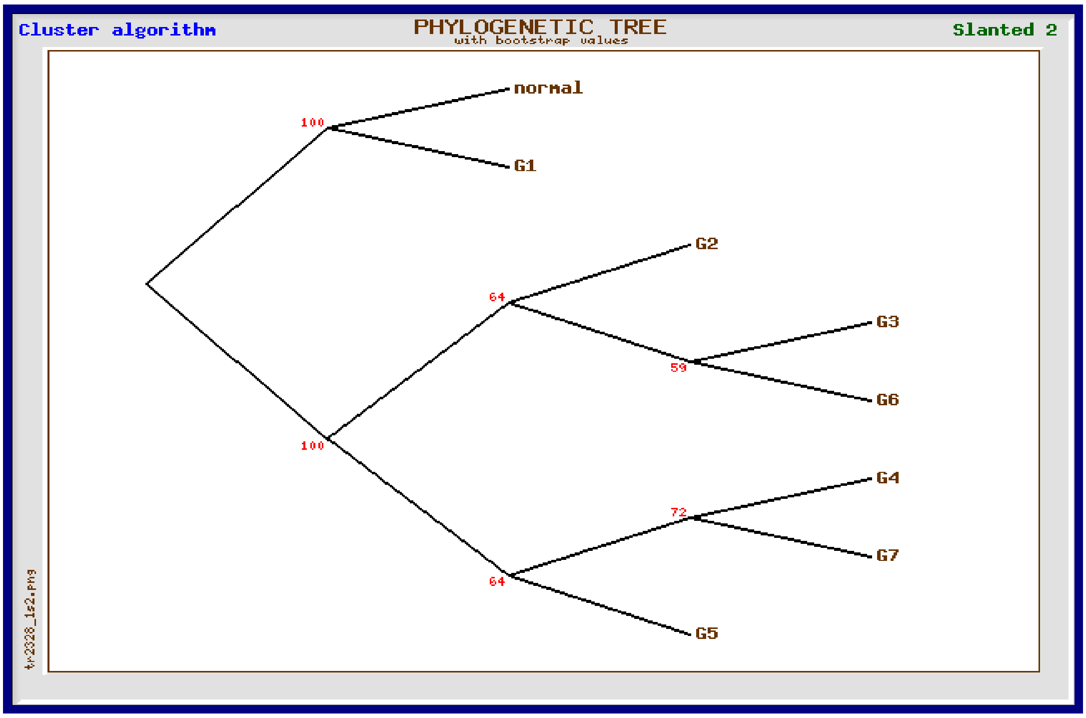


Fig. (80): A cladogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the seven studied groups; drawn by GeneBee program (Slanted cluster algorithm)

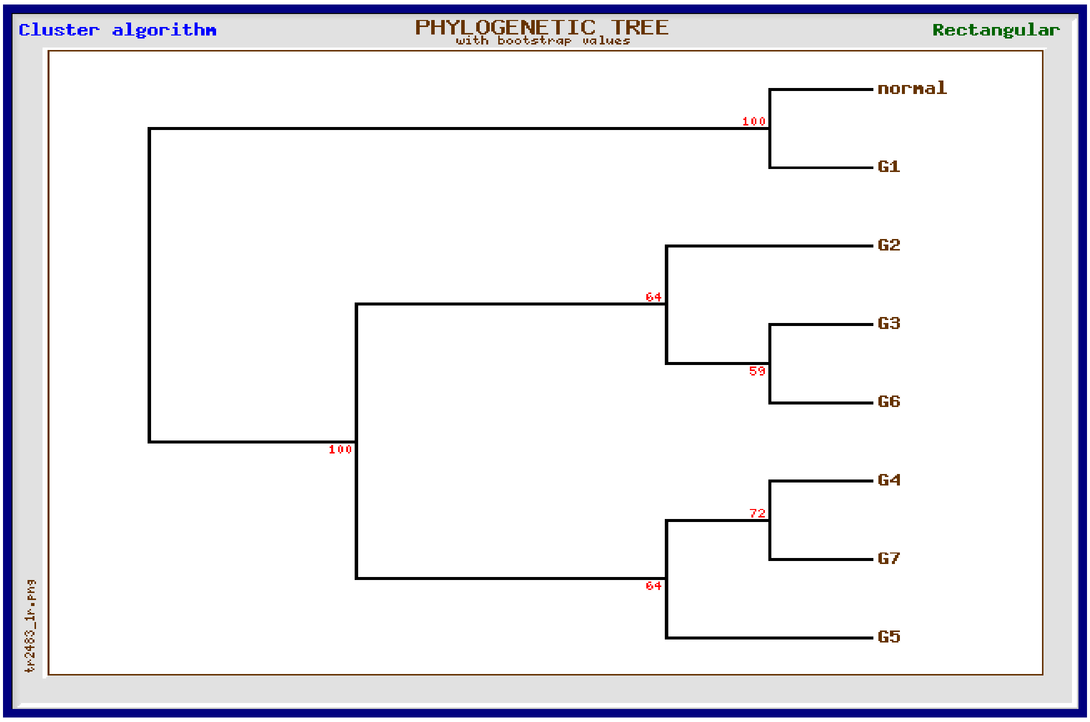


Fig. (81): A phylogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the seven studied groups; drawn by GeneBee program (Rectangular cluster algorithm)

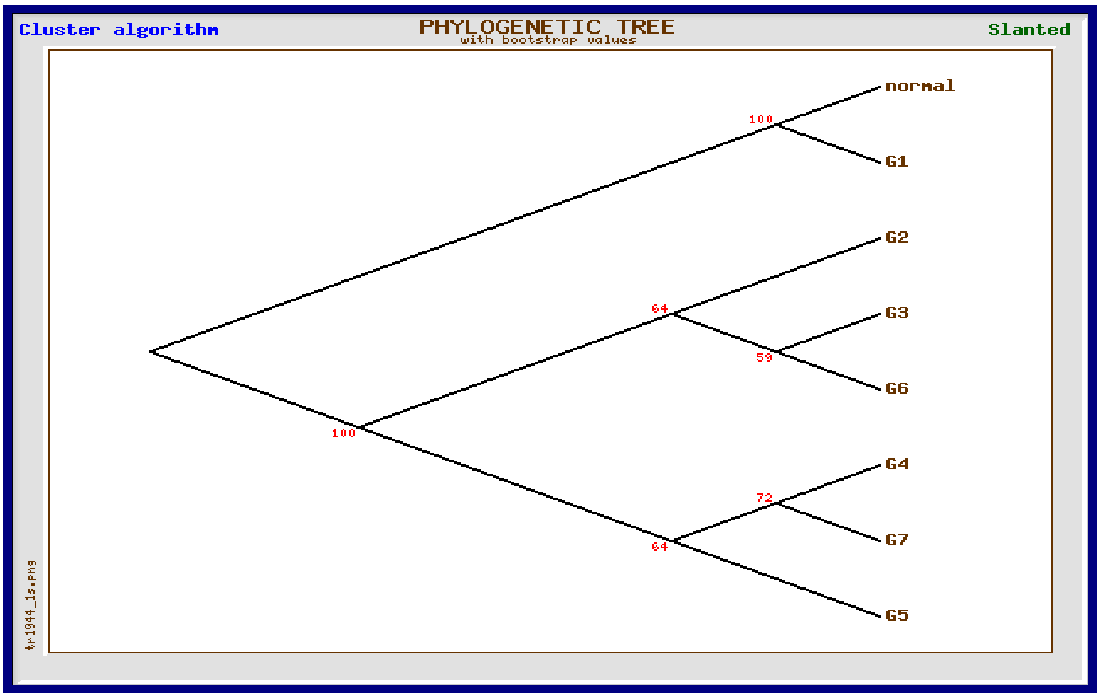


Fig. (82): A phylogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the seven studied groups; drawn by GeneBee program (Slanted cluster algorithm).

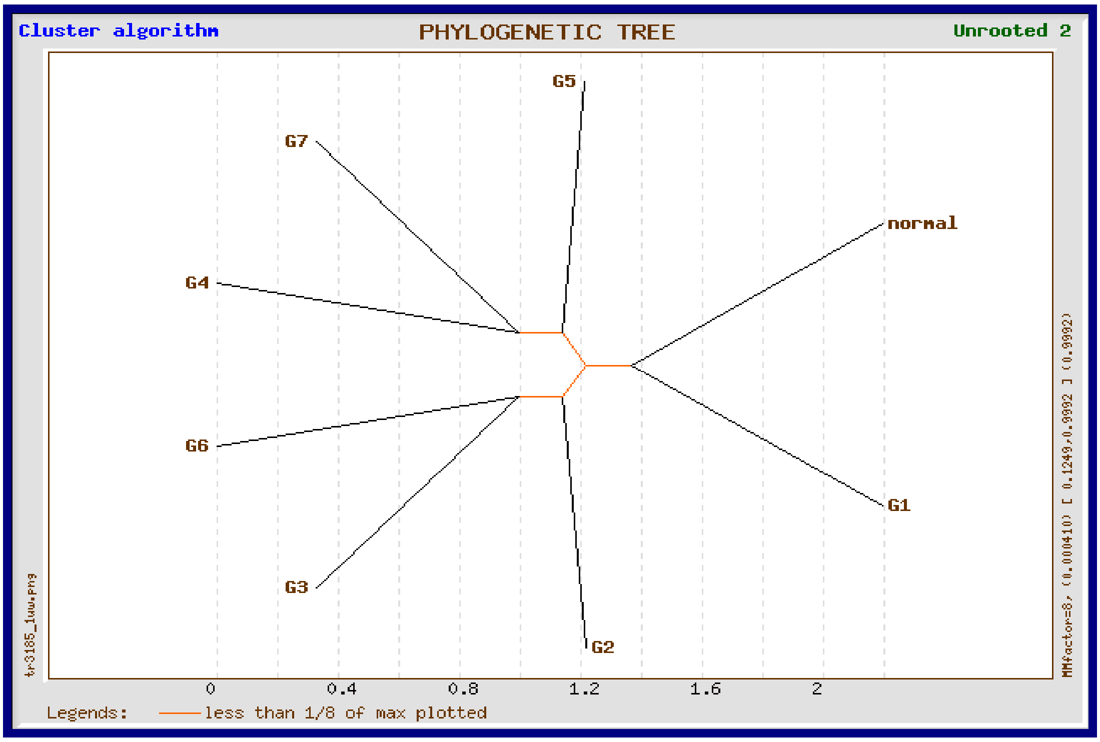


Fig. (83): A phylogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the seven studied groups; drawn by GeneBee program (unrooted cluster algorithm).

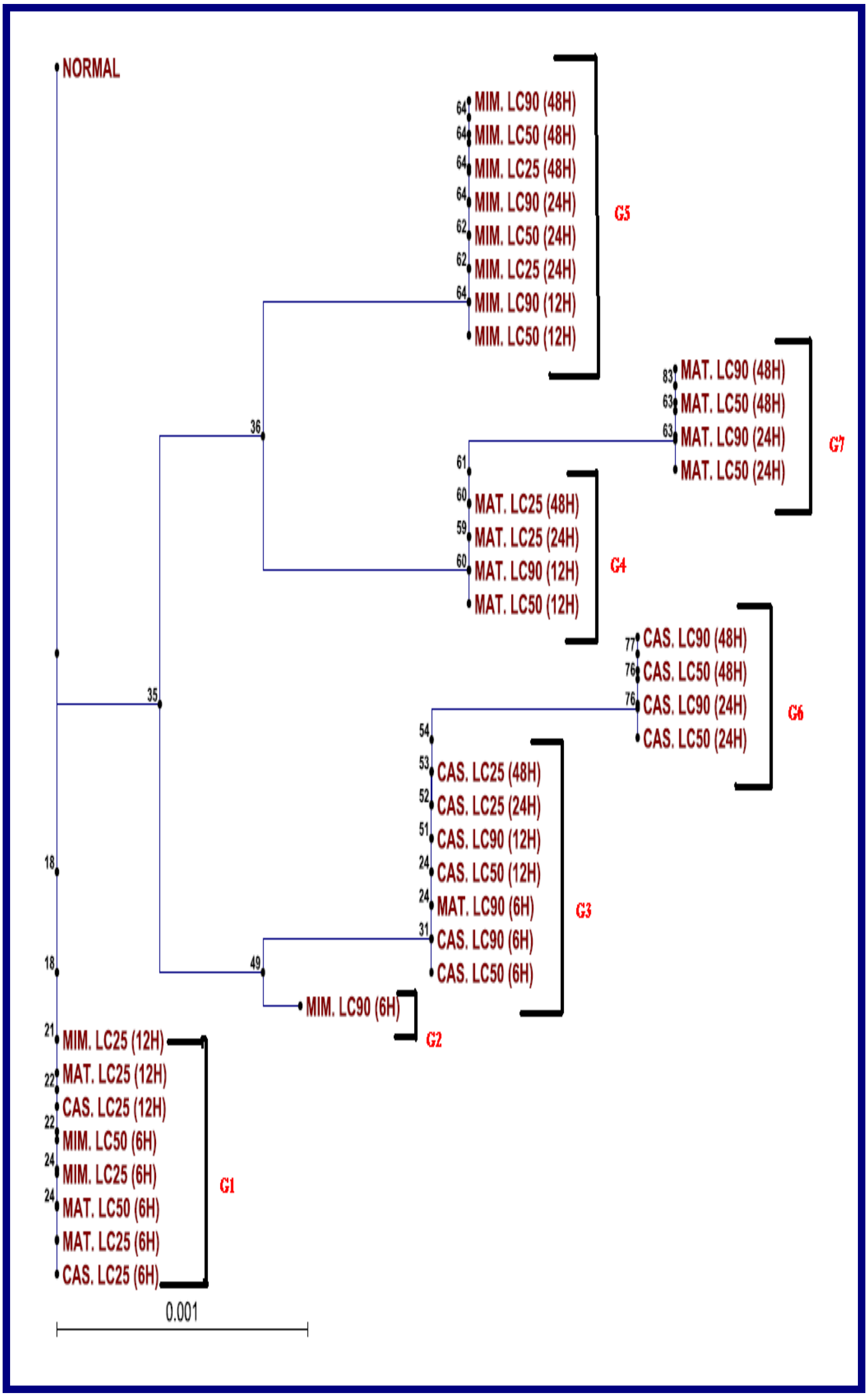


Fig. (84): A phylogram, with bootstrap confidence values at the nodes, based on the alignment of gene sequences of the studied doses.