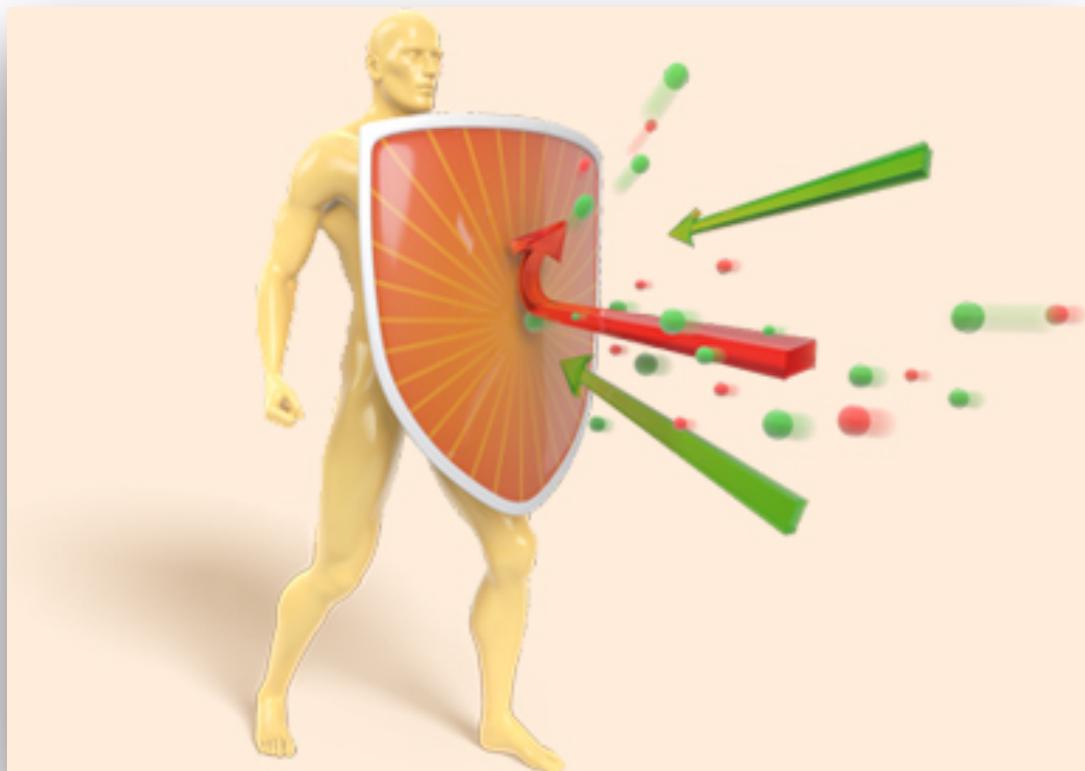


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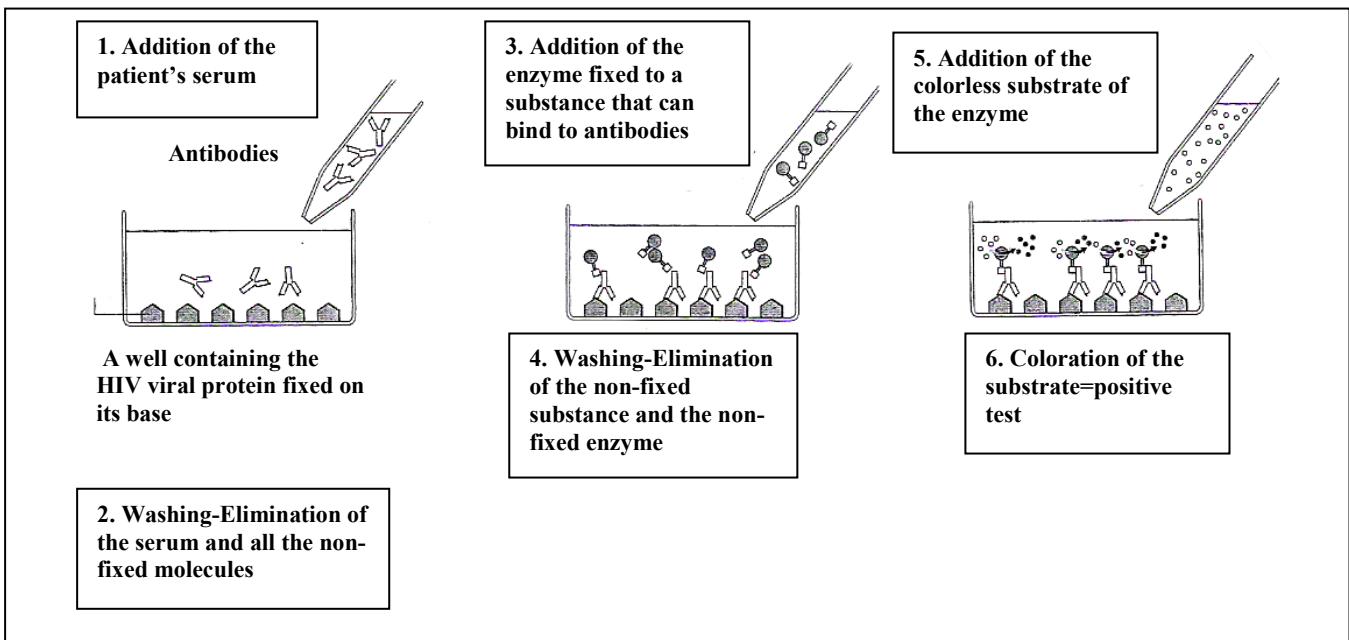
Official Exams 12 LS



Question III (6 pts.)

AIDS, or Acquired Immunodeficiency Syndrome is a disease due to a virus called HIV, or Human Immunodeficiency Virus. This disease affects the immune system and develops through many years, more or less rapidly depending on the individual.

Individual A is suspected to be infected by the virus. He consulted a doctor who prescribed blood analysis and a test called ELISA test. Document 1 reveals the different steps and the obtained results of this test.



Document 1. ELISA test: steps and result

- a- Write a short text describing document 1.
- b- What does the obtained result indicate? How can you explain this result?

Document 2 reveals the amount of T4 lymphocytes, measured over time, of a patient **B** who presents severe signs of infection.

Duration in months	3	6	12	18	30	40	50	70
Amount of T4 lymphocytes/mm ³ of blood	550	750	800	500	450	300	200	50

Document 2. Variation of the amount of T4 lymphocytes in function of time

- c- Draw the curve of the variation of the amount of T4 lymphocytes in function of time.
- d- Analyze the curve, then find out the cause of the observed immune deficiency starting from the 40th month.
- e- Knowing that the blood analysis done for patient A shows that the amount of T4 lymphocytes is equal to 800/mm³ of blood, and in reference to document 2, find out the duration of infection in patient A.

Question III (6 pts)

- a- We add the serum of the patient to a well containing the viral protein of HIV fixed on its base. Then we wash the well to eliminate the serum and all the non-fixed molecules. We then add an enzyme fixed to a substance capable to bind to antibodies. We wash again the well to eliminate the non-fixed substance and the enzyme. We finally add to the well a colorless substrate of the enzyme, a coloration appears indicating that the test is positive. (1 ½ pt)
- b- Patient A is seropositive. The positive test indicates that the serum of patient A contains anti-HIV antibodies. This means that individual A is infected and his immune system reacts to synthesize the specific antibodies. (½ pt.)
- c- (1 ½ pt.)

**Amount of T4 lymphocytes
/mm³ of blood**

T4 L/mm³ of blood

months

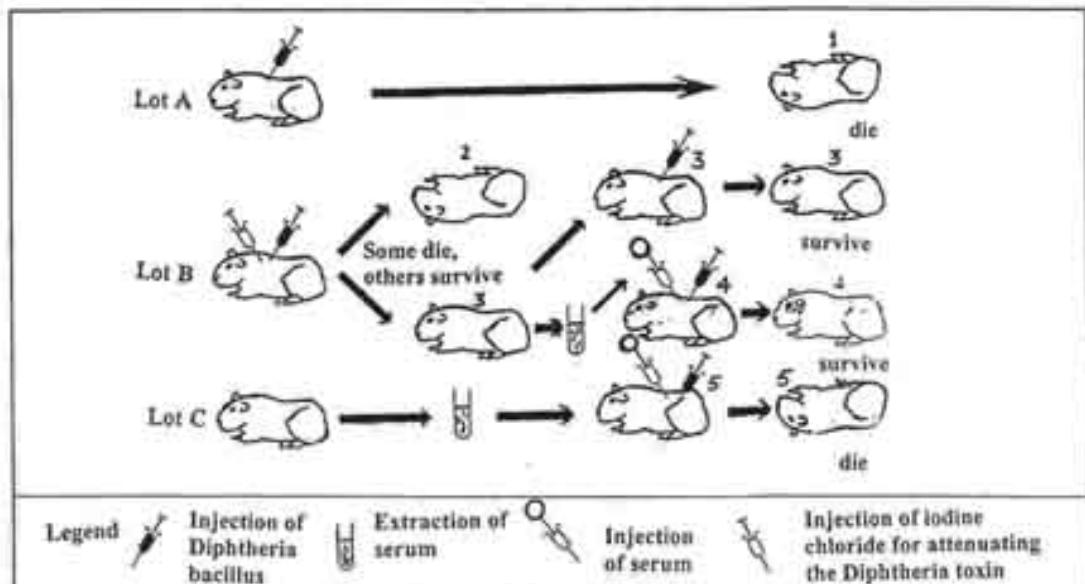
**Duration
(in months)**

Variation of the amount of T4 lymphocytes in function of time

- d- During the first 12 months, the amount of T4 increased from 550 / mm³ of blood to a maximum of 800/mm³ of blood. Starting from the 12th month, the amount of T4 decreased to become 50/mm³ of blood after 70 months. (1 pt)
The total immune deficiency observed, takes place starting from the 40th month, is due to the absence of T4 (destruction). (½ pt)
- e- Since the number of lymphocytes is 800/mm³ of blood, we can say that the duration of the infection is almost 12 months. (1 pt)

Question IV (8 pts)

A- In the framework of studying the transmission of immunity against diphtheria, a human disease caused by a bacillus that secretes a deadly protein toxin, the following experiments are conducted on Guinea pigs. Document I shows the experimental procedure used and the results obtained.



Document 1

- a- Describe, in a short text, each experiment performed and its obtained result.
 - b- Interpret these experiments. What can you deduce?
 - c- Indicate the two medical applications that you can draw out from these experiments? Justify the answer.

B- To understand why some infectious diseases can infect an organism only one time during life, even when the same organism is confronted with the same pathogenic microorganism again, we perform the following experiment.

We inject a Guinea pig with an attenuated antigen X, and we measure the amount of plasma anti-X antibodies. After 50 days, when the amount of anti-X antibodies in the plasma becomes nearly nil, we inject the same Guinea pig again with antigen X and another antigen: antigen Y. We measure the amount of plasma anti-X and anti-Y antibodies. The results are shown in document 2.

Time (in days)	0	8	15	30	50	60	75	85	100
Amount of anti-X antibodies (in a.u.)	0	0	1	0.8	0.2	1.5	3	2.8	2.6
Amount of anti-Y antibodies (in a.u.)	-	-	-	-	0	0	1	0.5	0

Document 2

- d- Draw, on the same graph the curves showing the variation of plasma anti-X and anti-Y antibodies as a function of time, specifying on the graph the contacts with the antigens.
 - e- Analyze the variations of the amount of anti-X antibodies, document 2. Draw out the characteristics of the secondary immune response.
 - f- What do the results of antigen-Y injection confirm?

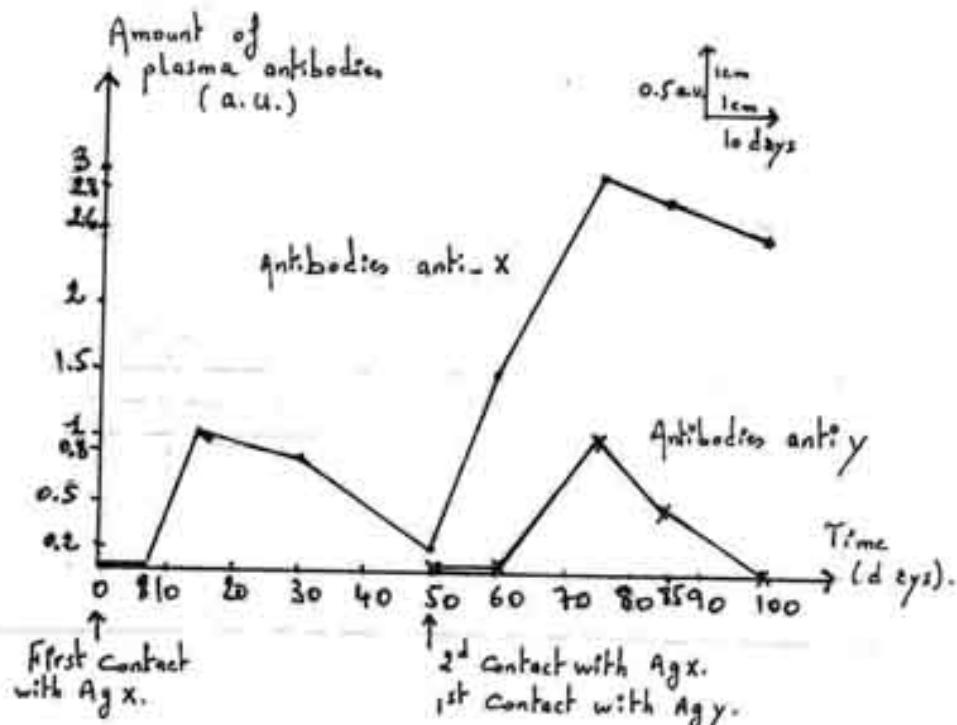
Question III (5 pts)

- a- II-4 and II-5: NN or Nd. Since they are phenotypically normal, each should have a dominant allele N and another allele that can be either N or d. (1 pt)
- b- II-4 and II-5 present the normal phenotypes. The risk for each of the two parents to be heterozygous is $2/3$ and the risk for two heterozygous couple to have an affected child is $1/4$, therefore, the risk of having a sick child is $2/3 \times 2/3 \times 1/4 = 1/9$ (1pt)
- c- Mutation has occurred at the site between 1.4 kb and 0.7 kb, because, the mutant allele, shows one fragment of 2.1 kb instead of two fragments 1.4 kb and 0.7 kb. (1 pt)
- d- II-4 has two of each fragment 1.4 kb, 1.2 kb, and 0.7 kb. These fragments correspond to the normal allele. Thus he is homozygous normal of genotype NN. (½ pt)
II-5 has one fragment 2.1 kb and 1.2 kb, which corresponds to the mutant allele, and fragments 1.4 kb, 1.2 kb, and 0.7 kb., which correspond to the normal allele. Thus, he is heterozygous normal of genotype Nd. (½ pt)
Fetus III-2 has a fragment 2.1 kb, which implies that he has received the mutant gene from his mother. He also has fragments 1.4 kb, 1.2kb, and 0.7 kb, which correspond to the normal allele that he received from his father. Thus, he will be normal heterozygous of genotype Nd. (½ pt)
- e- No, because the two parents are not heterozygous and the father II-4 who is homozygous gives only one type of gamete N. (½ pt)

Question IV (8 pts)

- a- We inject Guinea pigs of lot A with diphtheria bacillus, they die (1). We inject Guinea pigs of lot B with attenuated diphtheria toxin (iodine chloride + diphtheria bacillus), some Guinea pigs die (2) while others survive (3). We inject the Guinea pigs who survived (3) with diphtheria bacillus again, they survive. We extract serum from the surviving Guinea pigs (3) and we inject it into other Guinea pigs (4) together with an injection of diphtheria bacillus, they survive.
We extract serum from Guinea pigs of lot C and we inject them with diphtheria bacillus into other Guinea pigs (5), they die. (1 ½ pts)
- b- The injection of diphtheria bacillus (D.B), into Guinea pigs of lot A causes their death, while the injection of iodine chloride and D.B (attenuated diphtheria toxin) into Guinea pigs of lot B does not kill all the Guinea pigs, and those who survive (3) do not die even when they are injected with the D.B (3). Therefore, the attenuated toxin is not deadly, it causes immunity against diphtheria bacillus.
The injection of the serum of immunized Guinea pigs (3) into guinea pigs (4), not immunized, protects them from D.B, while the injection of serum from non-immunized Guinea pigs of lot C could not protect guinea pigs (5) against diphtheria bacillus. This means that the serum obtained from immunized Guinea pigs contains molecules that provides immunity against D.B. Thus, attenuated Diphtheria toxin provides immunity against diphtheria bacillus transferred by the serum.(2 pts)
- c- Vaccination and serotherapy. Because in the case of vaccination, we give an attenuated toxin, which allows the body to launch an immune response upon contact with the concerned antigen (Guinea pigs 3). In the case of serotherapy, we give the serum which contains antibodies, that are against the concerned antigens (guinea pig 4). (1pt)

d- (1 1/2 pts)



Variation of the concentration of the plasma anti-X and anti-Y antibodies

- c- The amount of plasma anti-X antibodies is nil at the beginning and starts to increase 8 days after the first contact with antigen X. It reached a maximum of 1 a.u on day 15 then it decreased progressively to reach 0.2 a.u on day 50. The second contact with the antigen X on day 50, causes a rapid increase in the amount of anti-X antibodies to reach a maximum of 3 a.u on day 75, then it decreases slowly to become 2.6 a.u on day 100.
Since the second contact with the same antigen causes the production of anti-bodies in larger quantity with less latent period and which persists longer, hence the secondary immune response is characterized by being more rapid, amplified and more persistent. (1 ½ pt)

- f. The results of antigen Y injection confirm that the immune response is specific, and at the first contact it is always slow, less amplified, and not persistent. (½ pt)

Question III (4 pts)

For determining the relation between the T4 lymphocytes and the T8 lymphocytes, also called cytotoxic T lymphocytes (Tc), we perform the following experiments:

- We remove from the spleen of a mouse, immune cells and we culture them in different mediums, document 1. We add to the culture mediums infected cells taken from an infected mouse of the same species. We detect cytotoxicity from the infected cells that are destroyed by the immune cells present in the mediums, document 2.

Medium 1	Immune cells in serum
Medium 2	Immune cells in a medium that leads to the elimination of T4 lymphocytes
Medium 3	Immune cells in a medium that leads to the elimination of T8 lymphocytes

Document 1

- a- Interpret the experiments done. What can you deduce concerning the condition for the appearance of cytotoxicity in a medium?

Experiment 1	Immune cells removed from medium 1 + infected cells from a mouse of the same species	Presence of cytotoxicity
Experiment 2	Immune cells removed from medium 2 + infected cells of a mouse of the same species	Absence of cytotoxicity
Experiment 3	Immune cells removed from medium 3 + infected cells from a mouse of the same species	Absence of cytotoxicity
Experiment 4	Immune cells removed from mediums 2 and 3 + infected cells from a mouse of the same species	Presence of cytotoxicity

Document 2

- The following microscopic observations reveal the mode of action of Tc lymphocytes in the presence of infected cells.

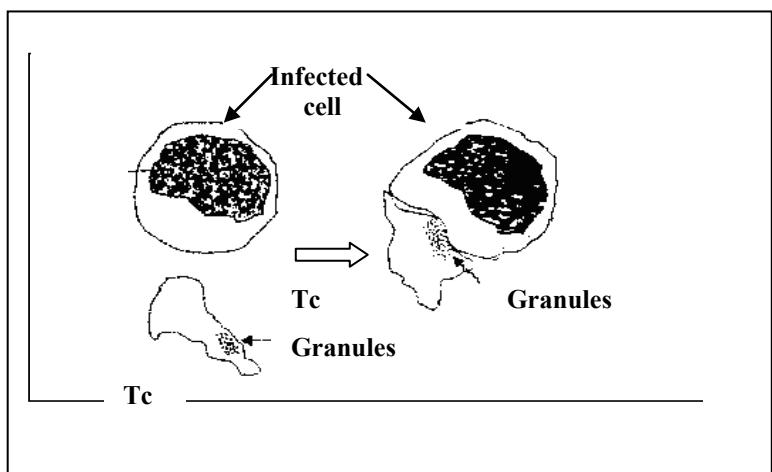
1st observation: In the presence of infected cells, the Tc lymphocytes that are rich in granules containing perforin, come in contact with these cells, document 3.

2nd observation: In the presence of non-infected cells, the Tc lymphocytes do not reveal granules containing perforin in their cytoplasm, and do not come in contact with these cells.

3rd observation: The membrane of the infected cells shows many pores in the region of contact with the Tc lymphocytes.

4th observation: In some mutant mice the Tc lymphocytes present a deficiency in perforin. No pores are observed at the level of the membrane of the infected cells in the region of contact with Tc lymphocytes, and the consequence is the non-destruction of the infected cells.

- b- Draw out from the analysis of the microscopic observations the role of perforin in the destruction of infected cells.
- c- From what has been preceded and based on the acquired knowledge, explain how the T8 lymphocytes become active cytotoxic T lymphocytes and how do they destroy the target cells.

**Document 3**

Question III (4 pts)

- a- In experiment 1 where cytotoxicity is observed, all the immune cells are present. On the other hand, cytotoxicity does not appear in absence of T4 (experiment 2) inspite of the presence of T8 and neither in absence of T8 (experiment 3) although T4 is present, which is confirmed by experiment 4 where cytotoxicity is observed where the immune cells taken from mediums 2 and 3 are placed together with the infected cells. This indicates that T4 only or T8 only are incapable to provoke cytotoxicity, thus, the presence of both is obligatory **(1 pt)**. Hence, the appearance of cytotoxicity necessitates the cooperation between T4 and T8. **(½ pt)**
- b- The microscopic observations reveal that in the presence of infected cells a contact takes place between Tc rich in perforin and these cells (1^{st} observation) while in the presence of non-infected cells, the Tc do not show perforin and are not in contact with these cells (2^{nd} observation). On the other hand, there is the appearance of pores in the region of contact between Tc and the infected cells (3^{rd} observation) and these pores do not appear in the case of a deficiency in perforin (4^{th} observation), Tc are thus, incapable to provoke the destruction of infected cells. From what has preceded, we can say that perforin is necessary when there is a contact between immune cells and infected cells, which is responsible for the formation of pores at the level of the membrane of the infected cells, followed by their destruction. **(1 ½ pt)**
- c- After recognizing the antigen, the activated T4 multiply and differentiate into cells that secrete interleukins. Interleukin 2 acts on certain T8 lymphocytes provoking their multiplication and their differentiation into effector cells: Tc lymphocytes. Tc binds to infected cells and secretes perforin that provokes the appearance of pores on the membrane of the infected cells. These pores permit the passage of granzymes that attack the DNA of the infected cells leading to their destruction. **(1 pt)**

Question II (5pts)

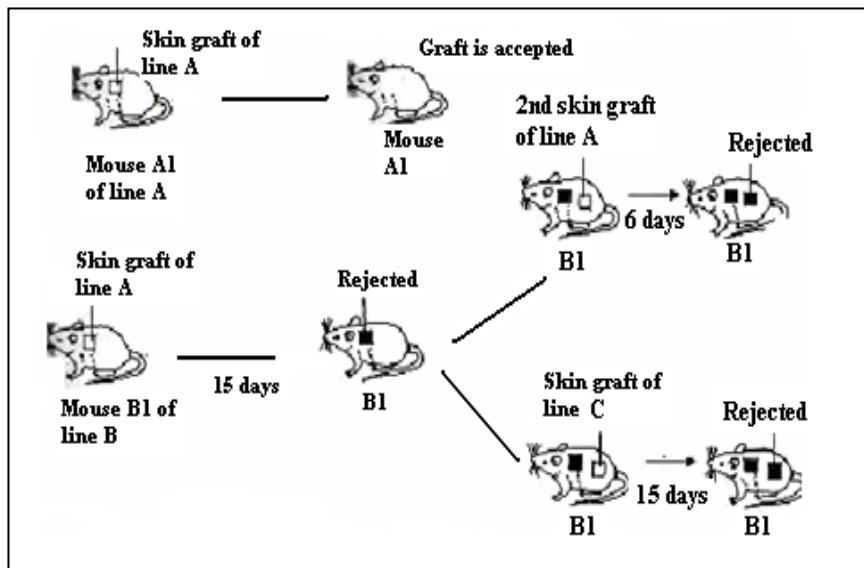
In the framework of studying immune responses, we do the following experiments.

1st Series of experiments: We perform skin graft between mice A, B, and C of different lines, document 1.

- a- Interpret these experiments.
- b- Indicate two characteristics of the immune system revealed by these experiments.

2nd Series of experiments: We graft skin of mouse A into mouse B under different conditions. The experiments done and the obtained results are shown in document 2.

- c- Starting from the analysis of document 2, specify the organs involved in graft rejection.

**Document 1**

3rd Series of experiments: We remove cells infected by virus X from a mouse of line A. We incubate the cells with radioactive chromium ^{51}Cr . This ^{51}Cr is absorbed and binds to proteins in the cells. After incubation, we wash these cells and culture them with different effector cells obtained from the same mouse A. The supernatants are then collected for measuring the quantity of ^{51}Cr released by the lysed target cells. Document 3a shows the experiments that are carried out, and document 3b shows the obtained results.

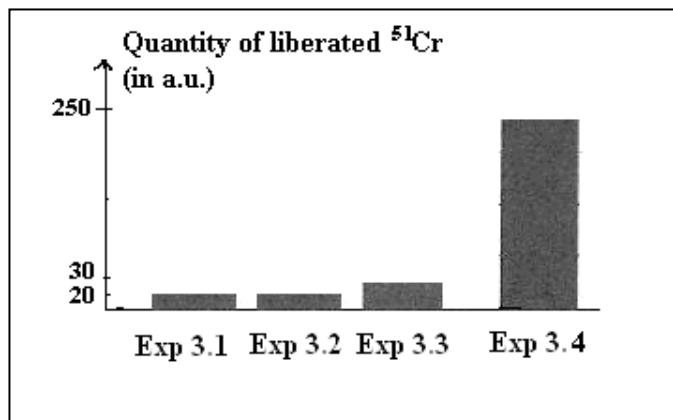
N° of experiment	Experimental conditions	Results
1	Control mouse B	Graft is rejected
2	Mouse B deprived of its thymus	Graft is accepted
3	Irradiated mouse B (destruction of bone marrow)	Graft is accepted

Document 2

N° of experiment	Effecter cells of mouse A
3.1	None
3.2	Macrophages
3.3	LT4 + LT8
3.4	LT4 + LT8 + macrophages

Document 3a

- d- Interpret the obtained results.
- e- By referring to the acquired knowledge, explain how the effector cells of document 3a intervene in the lysis of infected cells.

**Document 3b**

Question II (5pts)

- a- The skin graft of mouse A received by mouse A₁ of the same line is accepted while the skin graft of a mouse A received by mouse B₁ of line B different from A, is rejected after 15 days. This same mouse rejects a second graft of A, 6 days after grafting, on the other hand it takes 15 days to reject the graft received from a mouse of line C.
Therefore, the graft succeeds only between mice of the same line and the rejection of the graft is faster upon second recognition. **(1pt)**
- b- Recognition of the non-self by the immune system, the presence of an immune memory, and specificity of the immune response **(½ pt)**
- c- Skin graft taken from a mouse of line A to mouse B (control) leads to the rejection of the graft. On the other hand , the graft is accepted when it is done on mouse B that is deprived of its thymus (experiment 2) or on mouse B that is subjected to the irradiation of the bone marrow (experiment 3). Therefore, the thymus and the bone marrow are involved in graft rejection. **(1pt)**
- d- The quantity of ⁵¹Cr released by the cells lysed in a medium deprived of effector cells of mouse of line A and in a medium containing macrophages is 20 a.u. This quantity increases to become 30 a.u. in a medium containing LT4 and LT8 and reaches 250 a.u. in a medium containing LT4, LT8 and macrophages.
Hence, graft rejection requires the co-operation between these three types of immune cells. **(1pt)**
- e- The macrophages digest the free virus, recognized as non-self, and transform them into peptides and present them on HLA molecules of class II. These macrophages are thus, antigen presenting cells (APC). These latter migrate towards the lymphatic ganglia where they activate the LT4 (LT_H) that secrete IL-2.
IL-2 activates the LT8 (LT_c), which adheres to the membrane of the target cell and releases perforin and granzymes that perforate the membrane and degrades the DNA of the target cell leading to its lysis. **(1½ Pt)**

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مسابقة في علوم الحياة
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Answer the following exercises.

Exercise 1 (5pts)

To determine the cause of juvenile diabetes in humans, the following experiments were carried out on mutant rats of the same strain, in which diabetes appears in the first few months of their life.

1st experiment: 100 newborn mutant rats were brought, and divided into two lots, lot A and lot B. Lot A was subjected to the ablation of the thymus, the organ where T lymphocytes undergo maturation, and lot B was used as control. A few months later; the number of the rats that presented diabetes was determined, document 1.

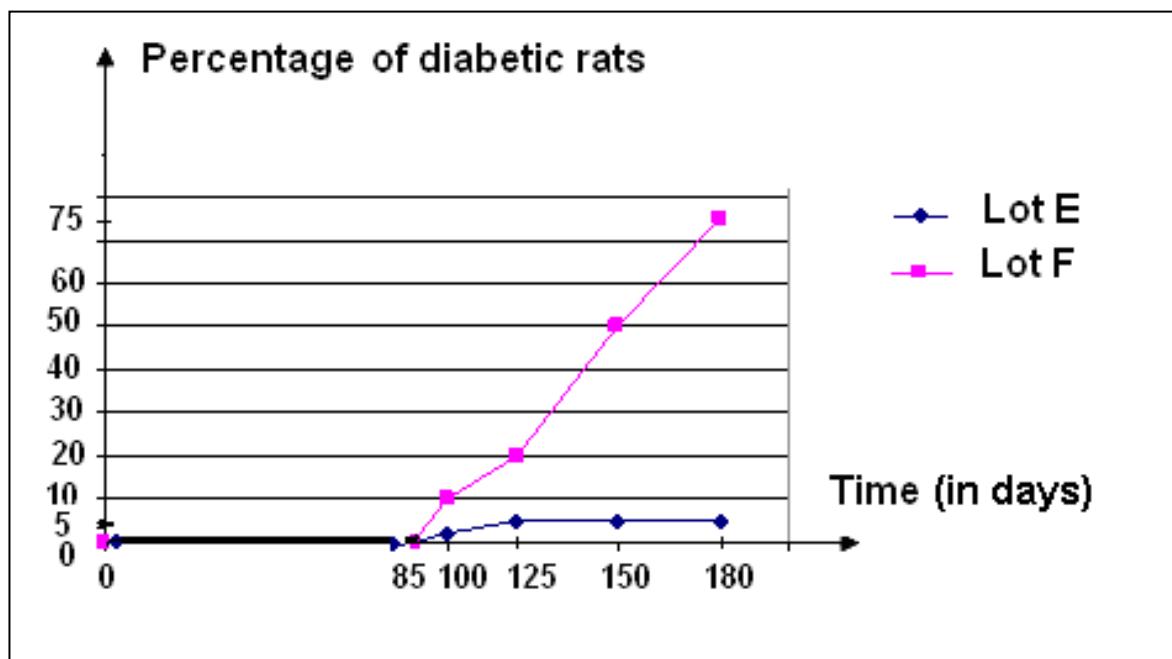
	Number of diabetic rats
Lot A	5/50
Lot B	30/50

Document 1

2nd experiment: Two lots of healthy non-mutant rats, lot C and lot D, were brought. The rats of lot C were injected with TL taken from diabetic mutant rats, and the rats of lot D were injected with TL taken from healthy rats. The rats of lot C, only, developed diabetes.

- 1- Formulate the hypothesis at the origin of these experiments.
- 2- Interpret each of the carried out experiments. What can one deduce regarding the formulated hypothesis?
- 3- What name can be attributed to this kind of disease? Justify the answer.

3rd experiment: Two lots of mutant rats, lot E and lot F, were brought. Lot E was treated, from birth, with cyclosporine, an immunosuppressant medicine, and lot F was used as control. Document 2 reveals the percentages of diabetic rats in these two lots of rats.



Document 2

- 4- Present in a table the different data provided by document 2.
- 5- Interpret the obtained results. Draw out the mode of action of cyclosporine.

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Exercise 1 (5pts)

- 1- Hypothesis: T lymphocytes of the mutant rats are at the origin of juvenile diabetes. **(0.5pt)**
- 2- The number of the rats that have juvenile diabetes is 5/50 in lot A, which had undergone ablation of the thymus. On the contrary, the number of the rats that have juvenile diabetes is larger (30/50) in the control lot B. This indicates that the thymus, the place of maturation of T lymphocytes, is implicated in the appearance of diabetes.
The 2nd experiment revealed that the healthy rats of lot C, injected with TL taken from mutant rats, developed diabetes, whereas the healthy rats of lot D, injected with TL taken from healthy rats, did not develop diabetes. This implies that the appearance of the disease is linked to the presence of the TL of the mutant rats. Thus the formulated hypothesis is valid and they are the TL of the mutant rats that are responsible for the disease. **(1.5pt)**
- 3- Auto-immune disease. **(0.25pt)**
The T lymphocytes are directed against the self; they recognize it as modified self and attack it. **(0.25pt)**

4- (1pt)

Time (in days)		0	85	100	125	150	180
Diabetic rats (in %)		0	0	2	5	5	5
Lot E		0	0	2	5	5	5
Lot F		0	0	10	20	50	75

Variation of the percentages of diabetic rats as a function of time in lots E and F

- 5- The percentage of diabetic rats was null in the two lots of rats until day 85. At day 100 the percentage increased to become 2% in the rats treated with cyclosporine, lot E, and 10% in the untreated rats, lot F. The percentage continued increasing to become 5% in lot E, and 20% in lot F. This percentage remained stable at 5% in the treated rats from day 125 to day 180, whereas it continued to increase in the untreated rats after day 125 to become 75% at day 180. This implies that the treatment with cyclosporine had prevented the appearance of diabetes in the mutant rats of lot E. Thus cyclosporine is a medicine that inhibits the action of TL responsible for the appearance of diabetes in the mutant rats. **(1.5pts)**

Exercise 3 (5pts)

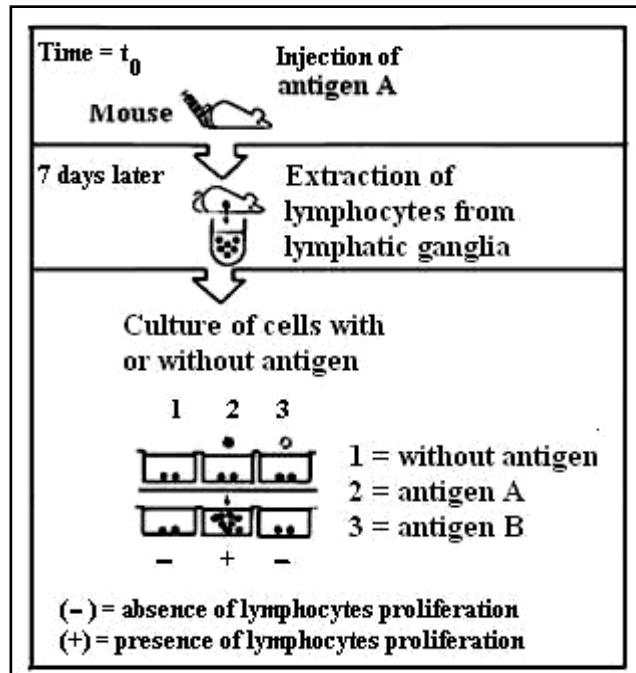
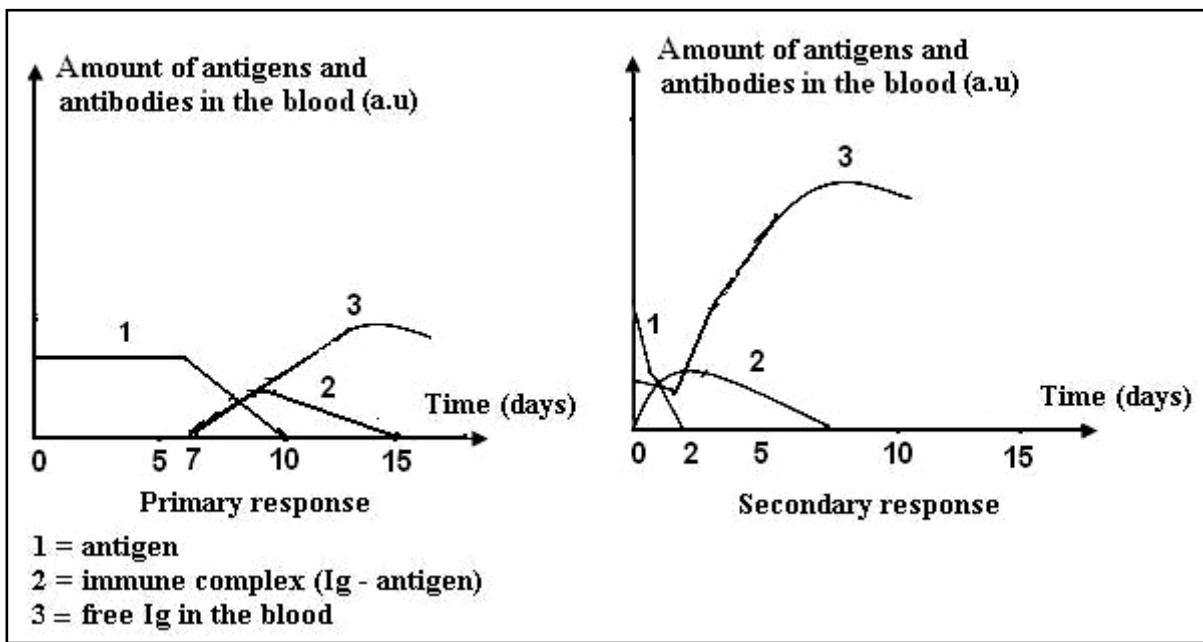
In order to study one of the characteristics of the immune response, the experiment shown in document 1 were carried out.

- 1- Write a short text describing the experiment carried out as well as the results obtained.
- 2- Interpret the obtained results. Draw out the characteristic of the studied immune response.

In a second experiment, the same steps are repeated without a seven days time delay. The cells of the lymphatic ganglia are directly extracted after the mouse immunization against antigen A. The results do not show any proliferation of lymphocytes.

- 3- Explain the necessity of the seven days time delay for the lymphocytes proliferation.

In a third experiment, we estimate the variations in the amounts of antigens and the produced antibodies (Ig) during two separate injections of the same antigen to an individual. The results are shown in document 2.

**Document 1****Document 2**

- 4- Compare the variations in the amounts of antigens then in the amounts of antibodies during both contacts. Deduce the characteristics of the immune memory.
- 5- Explain the appearance then the disappearance of the immune complexes following the antigen's injection.

Exercise 3 (5pts)

1- Antigen A is injected into a mouse. 7 days later, cells of the lymphatic ganglia are extracted and put in 3 culture mediums: without antigens in medium (1), with antigen A in medium (2), and with antigen B in medium (3). We observe the absence of lymphocytes proliferation in the 1st and 3rd culture mediums and the proliferation of lymphocytes occurs in the second medium. **(1pt)**

2- A high proliferation of lymphocytes extracted from the mouse immunized against antigen A was observed when they are put in culture with this antigen. On the contrary, no proliferation was observed when they are alone or in contact with antigen B. This implies that the proliferation of the lymphocytes, selected after the first contact with antigen A, cannot occur unless the lymphocytes are put again in contact with the same antigen. Thus the immune response is specific. **(1pt)**

3- “7 days time delay” is necessary to induce the immune response. Macrophages phagocytose the antigens and become APC that migrate towards the lymphatic ganglia. APC bind to the lymphocytes via their specific receptors and activate them. These selected lymphocytes rapidly multiply and proliferate upon a second contact with the same antigen. **(1pt)**

4- During the first contact with the injected antigen (primary response), the antigen's amount in the blood decreases starting day 7, to disappear within 10 days. However, during the secondary response, the antigen's amount in the blood decreases and disappears after 2 days, more quickly than at the time of the primary response.

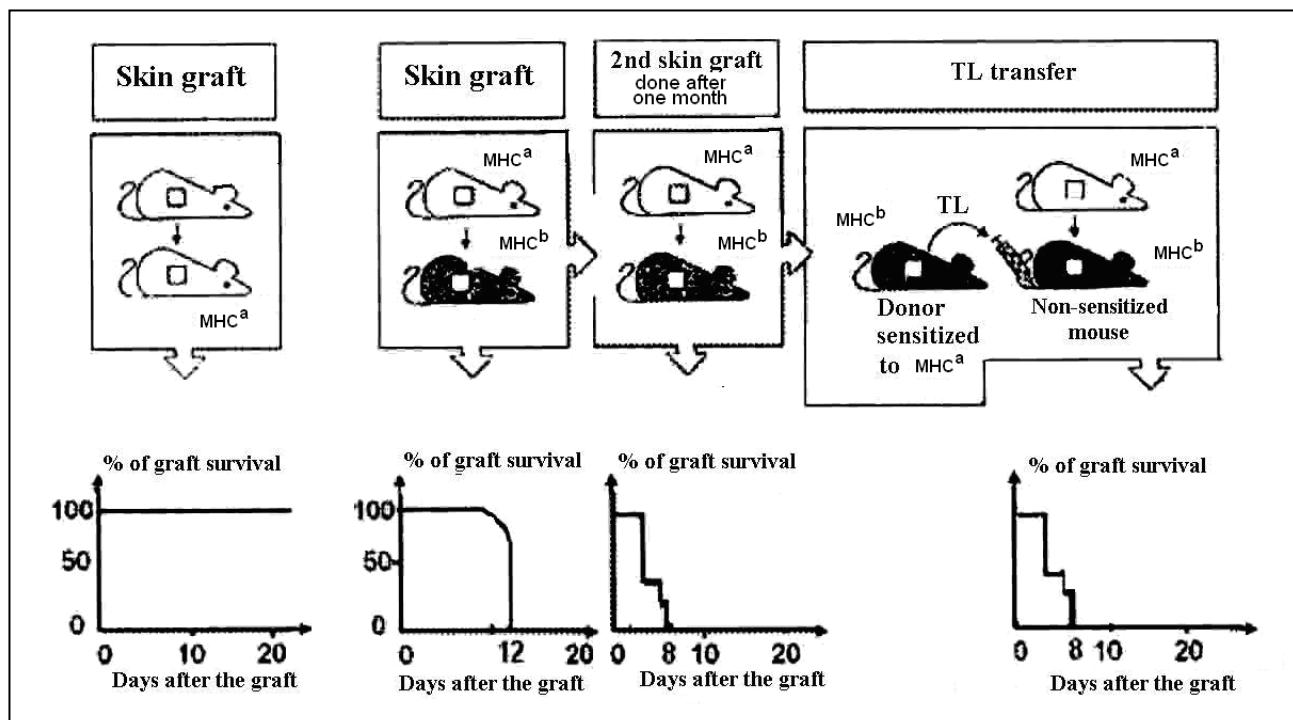
At the time of 1st contact with the injected antigen, the amounts of circulating antibodies in the blood are null and does not appear until the 7th day. They increase to reach a maximum at day 13. On the contrary, during the 2nd contact, these antibodies are present as of day 0, they start to increase at day 2, and reach a maximum at day 7, greater and faster than in the 1st contact. Beyond this day, the amount of antibodies in both cases decreases however remains higher in the 2nd contact.

This shows that during the 2nd contact the antibodies are produced earlier and in greater amount and the elimination of the antigens is faster. Thus the immune memory favors a faster, stronger, and more lasting response. **(1.5pt)**

5- The appearance of immune complexes is due to the neutralization of the antigen by the antibodies secreted by plasmocytes. The disappearance of these complexes is due to the opsonisation and phagocytosis carried out by macrophages. **(0.5pt)**

Exercise 3 (5 pts)

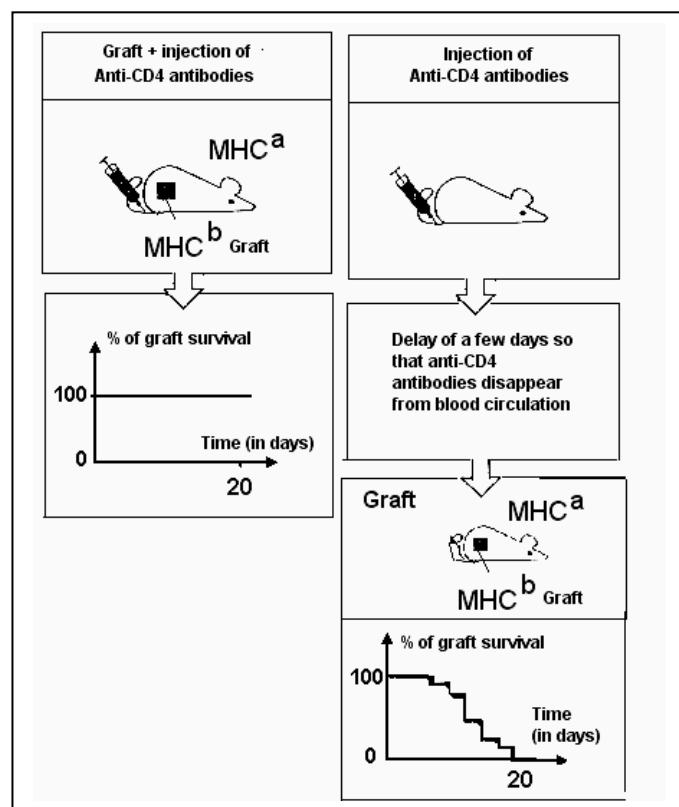
In order to know the mechanisms responsible for rejecting or accepting grafts, experiments on mice of the same line or different line are done. The experiments and their results are shown in document 1

**Document 1**

1. Interpret these experiments. Draw out the conditions of a graft rejection.

Two types of T-lymphocytes (TL) are recognized: TL4 with a CD4 receptor and TL8 with CD8 receptor. The experiments revealed in document 2 are carried out in order to determine the role of TL involved in graft rejection.

2. Interpret the obtained results. What can be deduced?
3. Justify that these experiments are not sufficient to assure, which of the two types of TL is involved in graft rejection.
Suggest an experiment that allows solving this problem.
4. Explain how the anti-CD4 antibodies intervene in accepting grafts. Draw out a practical medical application.

**Document 2**

Exercise 3 (5pts)

1. The skin graft from mouse MHC^a to another mouse of the same line shows a 100% survival of the graft that persists beyond 20 days. On the other hand, the graft carried out between two mice of different lines MHC^a and MHC^b survives 100% until day 10 after which the % of survival then decreases to become null, and the graft is rejected at the end of day 12. This % is even weaker when mouse MHC^b receives, after one month, a second skin graft from MHC^a and the graft is rejected on the 8th day < 12th day. The injection of TL, obtained from mouse MHC^b immunized against MHC^a, into a mouse MHC^b which receives a 1st skin graft from MHC^a, led to the rejection of the graft in 8 days as in the case of the 2nd graft.

This implies that graft survival can take place when it is done between members of the same line. Graft rejection is done between individuals of different lines and is faster after a 2nd contact with the same antigen and that TL are the cells that reject grafts.

Therefore, graft rejection done between two different lines necessitates the presence of TL. **(1.5pts)**

2. Graft between different lines, done along with the injection of anti-CD4 antibodies, succeeds and the percentage of its survival is maximal. On the other hand, if the injection of anti-CD4 antibodies is carried out a few days before the graft, a time delay for these antibodies to disappear from blood circulation, the graft is rejected. This means that the anti-CD4 antibodies, when they are present, prevent graft rejection. Therefore, TL4 cells have a role in graft rejection. **(1pt)**

3. This experiment is not sufficient to determine which type of cells is involved in graft rejection. Question 2 reveals that the TL4 cells have a role in graft rejection but it cannot be determined whether TL8 have the same role. **(0.5 pt)**

For that, it is necessary to repeat the experiments of document 2, and add two more mice. The 1st mouse is injected with anti-CD8 antibodies and the 2nd mouse with anti-CD4 and anti-CD8 antibodies, before performing the graft between the two different lines. We follow the variation of the graft: survival or rejection in order to determine which cell is behind graft rejection, TL4 or TL8 or both. **(1pt)**

4. Antibodies specific for CD4 are simultaneously injected with the grafting. They fix to TL4 receptors to block them. The blocked TL4 are not activated and do not proliferate nor do they differentiate into cells that secrete interleukins 2. Thus, the TL8 are not activated and the graft is successful. **(0.75 pt)**

Anti-CD4 antibodies can be used as immunosuppressor drugs during grafting. **(0.25pt)**

Exercise 2 (5 pts)

In an attempt to understand how the HIV that causes AIDS infects selectively T4 cells, we perform the following experiments on many lots of T4 cells (Lymphocytes characterized by the presence of CD4 proteins on their membranes) and T8 cells (lymphocytes characterized by the presence of CD8 proteins on their membranes).

Document 1 presents the experimental procedure and the obtained results.

1-Interpret the obtained results.

Studies and knowledge of the immune system and the immune reactions of persons infected by HIV allow for the preparation of an anti-HIV vaccine. We test the efficiency of this vaccine on Rhesus monkeys.

	Experimental Procedure	Results
Lot 1	T4 and T8 cells are placed directly in the presence of HIV	Infection of T4 cells, but no infection of T8 cells
Lot 2	T4 cells are incubated for 20 minutes with several types of antibodies* that do not bind to the membrane protein CD4, then placed with HIV	Infection of T4 cells only
Lot 3	T4 cells are incubated for 20 minutes with antibodies* that bind to the membrane protein CD4, then placed with HIV	No infection of T4 cells
*Anti-bodies block the biological activity of the molecules to which they bind		

Document 2 reveals the

Document 1

variation of the proportion of T8 cells specific to HIV during infection time in vaccinated and non-vaccinated monkeys.

	Time (in weeks)							
Proportion of T8 specific for the HIV (in a.u)	Exposure to HIV							
	0	1	2	4	6	8	10	12
Lot 1 : Vaccinated monkeys	0.1	7	6.5	6	4	3	2	2
Lot 2 : Non-vaccinated monkeys	0	0	0.5	2	1.5	1	1.3	1.5

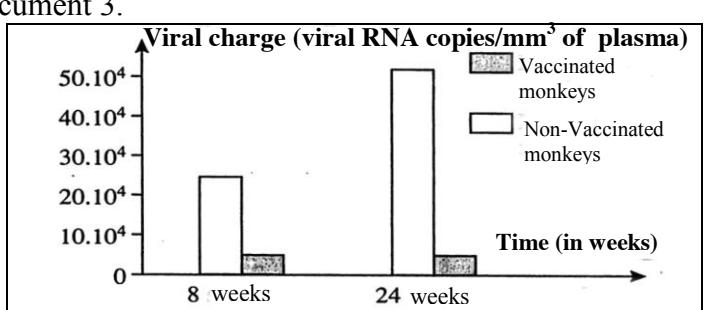
Document 2

2-Draw, on the same graph, the curves obtained from the tabulated data.

3-The immune response in the vaccinated monkeys is rapid and amplified. Refer to the results of document 2 to justify this affirmation.

We measure the viral charge (the number of viral RNA copies /mm³ of plasma that is an indicator of the concentration of the virus in blood) in the vaccinated and non-vaccinated monkeys after 8 and 24 weeks of exposure to the virus. The results are shown in document 3.

4-Compare the obtained results and draw out a relation concerning the effect of the studied vaccine.

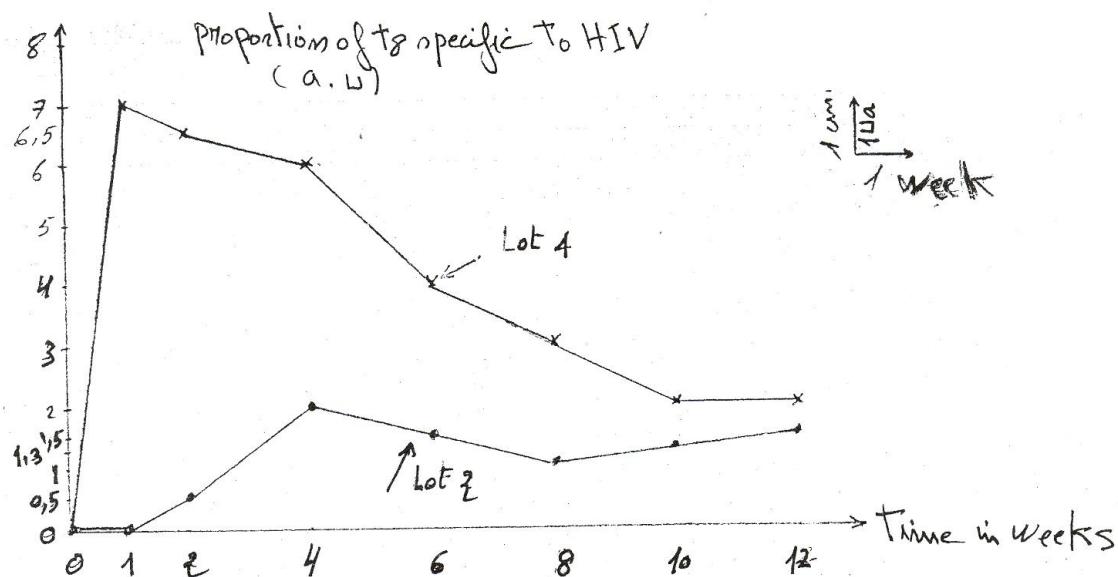
Document 3

Exercise 2 (5 pts)

- 1- The results of lot 1 show that T4 and not T8 cells get infected by HIV. This means that T4 are the target cells of HIV. Moreover, results show that infection of T4

cells occurs in lot 1 and lot 2 where CD4 proteins are free, while no infection of T4 cells occurs in lot 3 where the CD4 proteins are fixed to antibodies. This means that HIV attacks only T4 cells possessing free CD4 on their membrane.

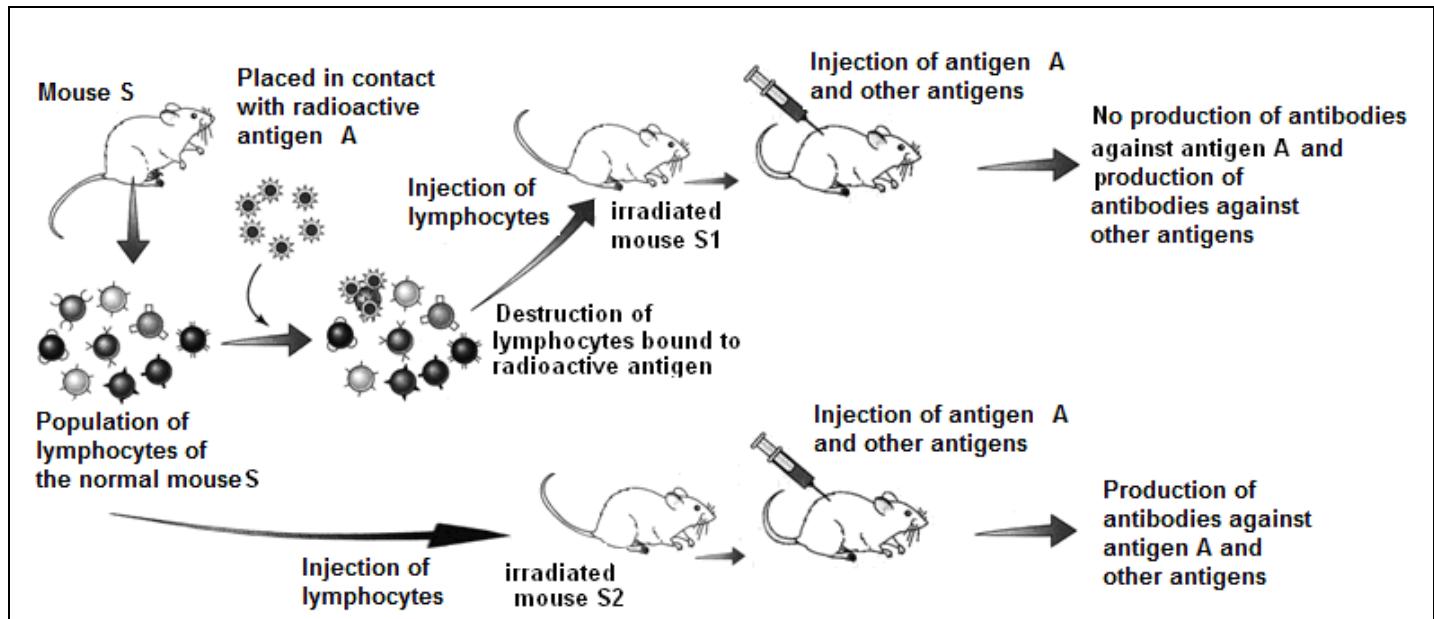
- 2- Graph showing the variation of the proportion of T8 specific to HIV in function of time in vaccinated and non vaccinated monkeys. (2 pts)



- 3- Document 2 reveals that in the vaccinated monkeys, the proportion of T8 specific to HIV start increasing from the time of viral exposure to reach 7 a.u. after one week while, in the non vaccinated monkeys, the proportion of T8 starts increasing after a longer period of time (two weeks) following infection. This explains why the immune response is rapid in the vaccinated monkeys. Also, document 2 shows that the proportion of T8 in the vaccinated monkeys reach a value 7 a.u which is higher than the value attained in the non vaccinated monkeys which is 2 a.u. so, the immune response in the vaccinated monkeys is amplified.
(1 pt)
- 4- In the non vaccinated monkeys, the viral charge is around $25 \cdot 10^4$ copies/mm³ of plasma after 8 weeks following viral infection and increases to reach $50 \cdot 10^4$ copies in mm³ of plasma after 24 weeks. These values are always higher than the viral charge in the vaccinated monkeys which remains constant at $5 \cdot 10^4$ copies/mm³ of plasma at weeks 8 and 24. (1 pt)

Exercise 3 (5 pts.)

In the framework of studying clonal selection of B lymphocytes which are at the origin of antibodies, we perform several experiments on mice of line S that are not immunized against an antigen A (document 1). According to the theory of clonal selection, each lymphocyte acquires during its development the ability to react with a specific antigen, even without being previously exposed to it.

**Document 1**

N.B. irradiation leads to the destruction of immune cells.

1- Refer to document 1 to show that:

- 1-1 B lymphocytes are ready to respond to an antigen before encountering it;
- 1-2 B lymphocytes possess a surface receptor;
- 1-3 The immune response is specific.

To determine the phenomenon responsible for the secretion of the most effective antibodies, researchers performed the following experiment. They injected mice with a chemical substance recognized by the immune system as a foreign antigen. This antigen is characterized by having several antigenic determinants.

At different times following the injection, researchers sacrificed the mice and dissected their lymphatic ganglia to detect the B lymphocytes which recognize the injected antigen. Document 2 shows the results of this experiment.

Time since the injection of the antigen (in days)	Aspect of the lymphatic ganglia	Number of the different detectable B lymphocytes clones	Efficiency of the immune response
5	Beginning of swelling	10	Average
10	Strong swelling	1 or 2	Very high

Document 2

- 2- Explain the swelling of the lymphatic ganglia mentioned in document 2.
- 3- To what can we attribute the number of B Lymphocytes clones 5 days following the injection of the antigen?
- 4- Formulate a hypothesis that explains the decrease in the number of B lymphocytes clones detected 10 days following the injection of the antigen.

Exercise 3 (5 Pts)

- 1-1** B lymphocytes are ready to respond to an antigen before encountering it, because we observe anti-A antibodies production in mouse S2 which received all lymphocytes. On the contrary, there is no production of anti-A antibodies in mouse S1 which received all lymphocytes except the lymphocytes that can recognize antigen A (already destroyed by radioactivity after the fixation on the radioactive antigen A). This indicates that the lymphocytes that recognized antigen A were present before any contact with this antigen. **(1 pt)**
- 1-2** The experiment shows that the radioactive antigen is fixed on plasma membrane of the B lymphocytes that recognized this antigen. This implies the presence of a membrane receptor capable of the fixation of this antigen. **(1 pt)**
- 1-3** The immune response is specific to this antigen because we observe the production of antibodies against all antigens except anti-A antibodies in mouse S1 that received all lymphocytes except the lymphocytes that can recognize antigen A. **(1 pt)**.
- 2-** The swelling started after 5th day and the strong swelling observed on the 10th day correspond to the activation of lymphocytes(T4 and BL) and to their rapid and important proliferation on day 10 leading to the formation B lymphocytes clones that recognize the antigen. **(1 pt)**
- 3-** The 10 clones of BL 5 days following the injection is attributed to the presence of 10 different antigenic determinants at the level of the antigen. **(½ pt)**
- 4-** Hypothesis: There is an important clonal selection of B lymphocytes where only lymphocytes that can recognize the most effective antigenic determinant are kept. **(1/2 pt)**

Or

There is an important clonal selection of B lymphocytes where only lymphocytes that can recognize the most frequent antigenic determinant are kept.

Exercise 3 (5 points) Specificity of Lymphocytes and Antibodies

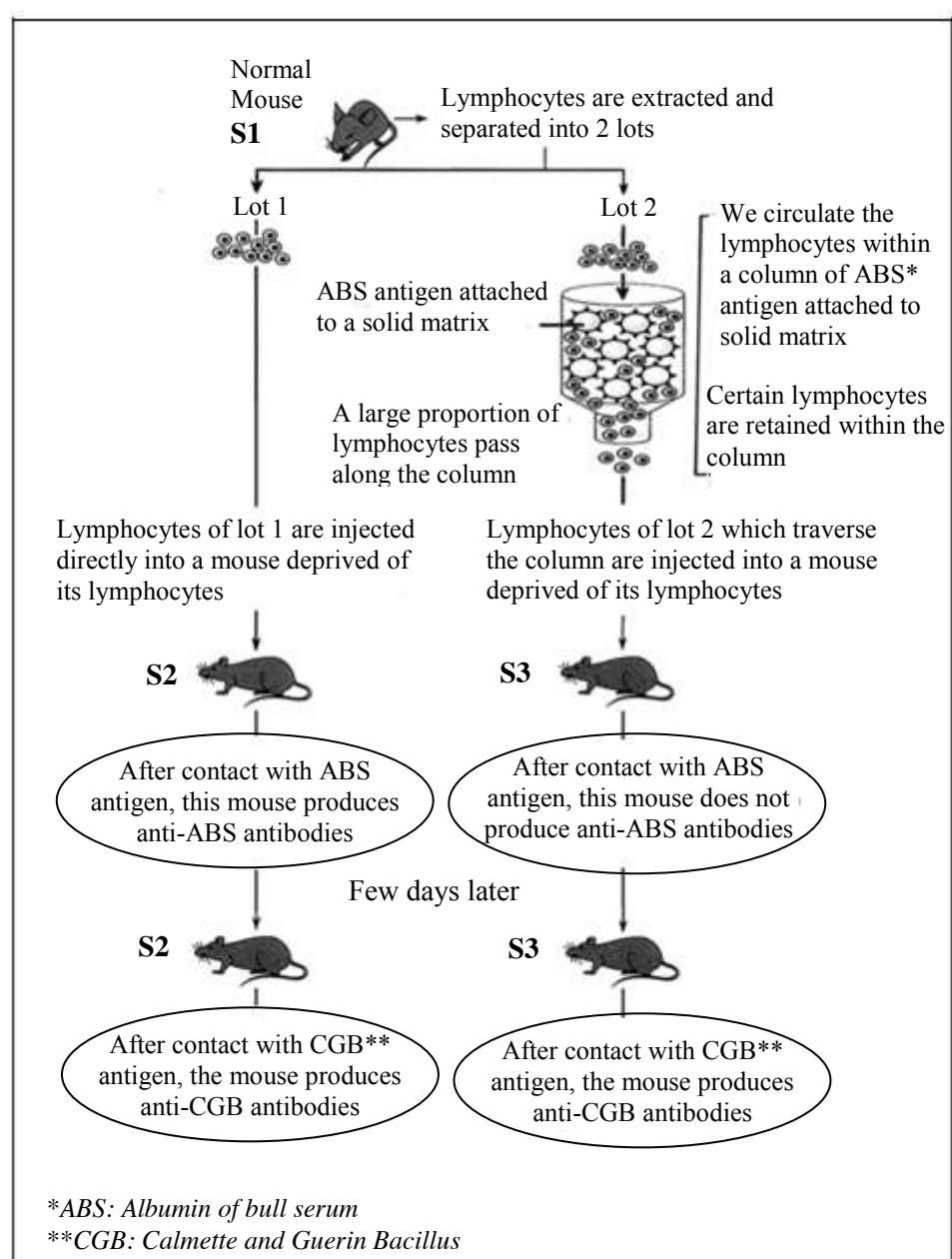
In the midst of the twentieth Century, two hypotheses were proposed to explain the high diversity of antibodies.

First hypothesis: Any lymphocyte encountering any antigen is capable of producing antibodies specific to this antigen.

Second hypothesis: Only some lymphocytes which correspond to an antigen are capable of producing antibodies specific to this antigen

To verify one of these two hypotheses, an experiment was performed on mice of the same strain. The steps and results of this experiment are represented in document 1.

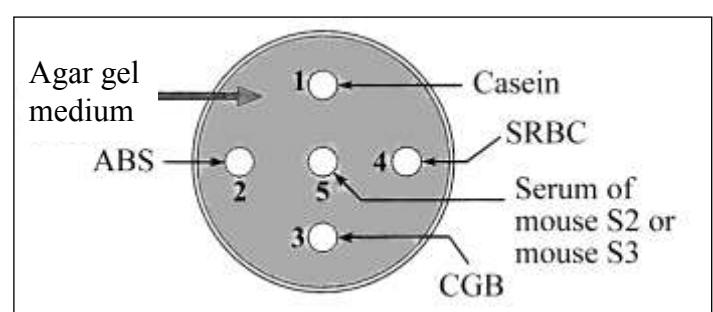
- 1- Write a text which describes the experiment shown in document 1.
- 2- Interpret this experiment to deduce which hypothesis is validated.
- 3- Name the different types of lymphocytes implicated in the immune response revealed by this experiment.



Document 1

Document 2 shows a serological test called, immuno diffusion in gel where antibodies and antigens are deposited in wells in agar gel medium. We deposit an antigenic substance in each of the wells 1, 2, 3 and 4 and deposit either serum taken from mouse S2 or serum taken from mouse S3 in the central well 5.

- 4- Specify where precipitation arc (s) would be formed with each serum after 24 hrs of antigens' deposit. Justify the answer.
- 5- Schematize the mechanism which leads to the formation of the precipitation arcs.

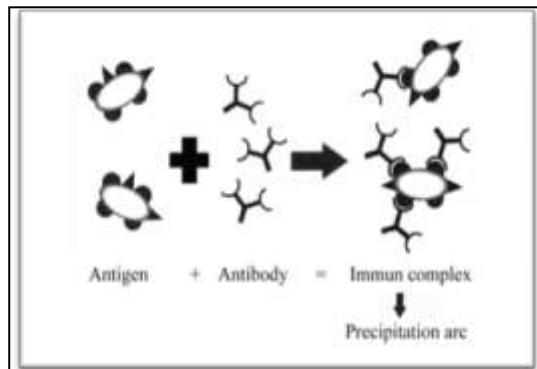


Document 2

Exercise 3 (5 points)

- 1- We extract lymphocytes from a normal mouse S1 and separate them into two lots. Lymphocytes of lot 1 are directly injected into mouse S2 deprived of its lymphocytes. After contact with ABS antigen (albumin of bull serum), this mouse produces anti- ABS antibodies. We circulated the lymphocytes of lot 2 in a column of solid matrix attached to ABS antigen. Some lymphocytes are retained within this column and a great proportion of these lymphocytes pass along the column. The lymphocytes of lot 2 which traverse the column are injected in mouse S3 deprived of its lymphocytes. After the contact with ABS antigen, mouse S3 doesn't produce anti- ABS antibodies. Several days later, following contact with CGB antigen (Calmette and Guerin Bacillus), mice S2 and S3 produce anti-CGB antibodies. ($1 \frac{1}{2}$ pt).
- 2- Mouse S2, deprived of its lymphocytes, and which receives lymphocytes extracted from a normal mouse S1, produces anti-ABS antibodies upon its contact with ABS antigen. On the contrary, no anti-ABS antibodies are produced by mouse S3 also deprived of its lymphocytes, and which receive lymphocytes that traverse a column of ABS antigens attached to solid matrix, upon its contact with the same antigen. This means that mouse S2 only receives lymphocytes specific to ABS antigen. ($\frac{1}{2}$ pt) However, these two mice S2 and S3 were able to produce anti- CGB antibodies upon their contact with CGB antigen. This means that both mice possess BL specific to CGB antigen. ($\frac{1}{2}$ pt). Thus, the production of specific antibodies, against a certain antigen, necessitates the presence of a category of BL specific to that antigen. **Therefore**, lymphocytes are specific to an antigen and this validates the second hypothesis. ($\frac{1}{2}$ pt).
- 3- B lymphocytes ($\frac{1}{4}$ pt) and T4 Lymphocytes ($\frac{1}{4}$ pt)
- 4- With the serum of mouse S2, a precipitation arc is formed between well 2 and well 5; and between well 3 and well 5 ($\frac{1}{4}$ pt), because mouse S2 produced anti -ABS antibodies and anti-CGB antibodies specific to ABS and CGB antigens respectively. ($\frac{1}{4}$ pt).
With serum of mouse S3, the precipitation arc is formed between well 3 and well 5 ($\frac{1}{4}$ pt), because mouse S3, in the absence of lymphocytes specific to ABS antigen, produces only anti-CGB antibodies which have an antigen binding site specific to CGB antigen. ($\frac{1}{4}$ pt).

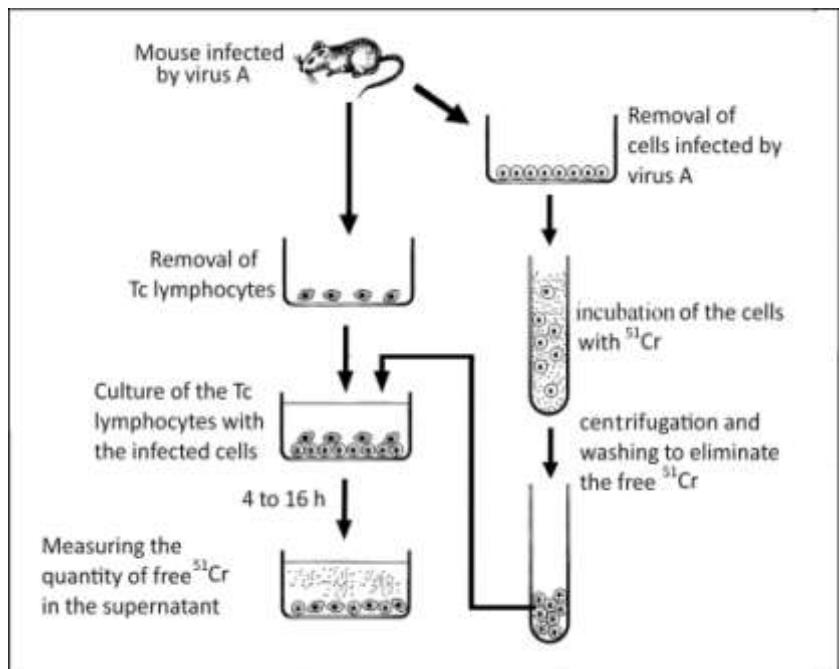
5- Drawing ($\frac{1}{2}$ pt)



Exercise 2 (5 points)**Cytotoxicity of Tc lymphocytes**

The cells that are infected by a virus express on their plasma membranes some antigens of this pathogen. These antigens can be recognized by specific receptors of the cytotoxic lymphocytes (Tc). In the attempt to prove the cytotoxicity of the Tc lymphocytes, we perform the experiment schematized in document 1, where cells infected by virus A are incubated with ^{51}Cr , a substance that binds to intracellular proteins after being absorbed by the cell.

1. Describe the experiment schematized in document 1.
2. Justify, by referring to document 1, how the presence of free ^{51}Cr in the supernatant reveals the cytotoxic role of Tc lymphocytes.
3. Specify the type of the immune response revealed by the experiment of document 1. Justify the answer.

**Document 1**

Dermal cells of mice of strain X or of strain Y, infected or not by a virus, are cultured in vitro. Tc lymphocytes removed from other mice of strain X, infected or not by a virus, are added to the culture medium. Document 2 presents the experimental conditions and the obtained results.

Origin of the cultured dermal cells Origin of the added Tc lymphocytes	Healthy mice X	Mice X infected by virus A	Mice X infected by virus B	Mice Y infected by virus B
Healthy mice X	No destruction of dermal cells	No destruction of dermal cells	No destruction of dermal cells	No destruction of dermal cells
Mice X infected by virus A		Destruction of the infected dermal cells by the Tc lymphocytes	No destruction of dermal cells	
Mice X infected by virus B		No destruction of dermal cells	Destruction of the infected dermal cells by the Tc lymphocytes	

Document 2

4. Interpret the results of document 2 and deduce the conditions that are indispensable for the functioning of the Tc.
5. Explain the mechanism that leads to the destruction of target cells by Tc lymphocytes.

Exercise 2 (5 points)

1. We remove from mouse X infected by virus A cells infected by virus A and Tc lymphocytes. We incubate the infected cells with ^{51}Cr , then we perform centrifugation and wash to eliminate the free ^{51}Cr . After that we culture the removed Tc lymphocytes with the infected cells. After 4 to 16 h we measure the quantity of free ^{51}Cr in the supernatant. **(1 ½ pt)**
2. The disappearance of ^{51}Cr after washing then its reappearance in the supernatant after culturing the infected cells with Tc lymphocytes indicate that these latter have destroyed the infected cells liberating the ^{51}Cr bound to intracellular proteins. **(1pt)**
3. Specific cell mediated immune response **(1/4 pt)**. Since the Tc cells have destroyed the infected cells. **(1/4 pt)**
4. Dermal cells of healthy mice are not destroyed by the Tc regardless from which mice they are removed.

Dermal cells of mice X infected by virus A or by virus B are only destroyed by Tc lymphocytes of mice X infected by the same virus: respectively virus A or virus B. Thus the cells infected by a virus are destroyed only by Tc of mice infected by the same virus. However dermal cells of mice of strain Y infected by virus B are not destroyed by Tc of mice of different strain X even though they are infected by the same virus B; thus the infected cells are destroyed only by Tc of mice of the same strain. **(1 pt)**

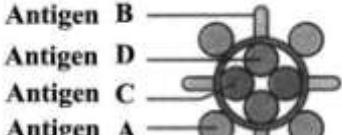
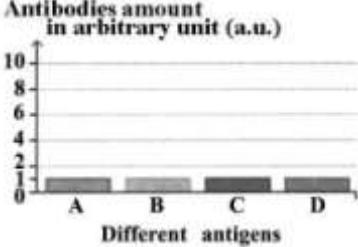
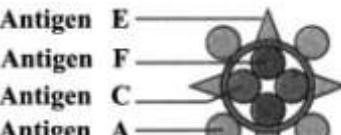
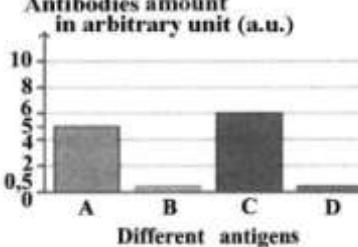
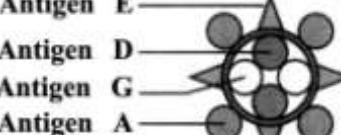
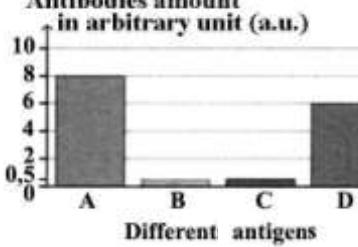
Therefore, the Tc cells destroy the infected cells if they belong to the same strain and are infected by the same virus. **(1/2 pt)**

5. The Tc lymphocytes recognize and bind, by its TCR, to target cells expressing the modified self: self MHC carrying a non self peptide of the antigen that is at the origin of their activation. They will then release, by exocytosis, perforin molecules forming hollow polyperforin channels through the cell membrane, and then they release granzymes molecules that penetrate the target cell through these channels leading to its DNA degradation and to the target cell destruction. **(1/2 pt)**

Exercise 2 (5 points)**Immunological memory**

When an organism encounters the same pathogen more than once during its lifetime, the immune response against this pathogen becomes more and more efficient. The flu virus exists in different variants having different antigens. We study the immune responses triggered by an individual upon contact with the flu virus three times during his life time.

The document below presents the age of this individual at the time of contact with one of the three variants of the flu virus and the evolution in the amount of antibodies specific to the antigens of variant 1.

Age of the individual at the time of infection	Antigens of the variant	Evolution in the amount of antibodies specific to the antigens of variant 1										
 2 years old individual in contact with variant 1 of the flu virus	 Variant 1 of the flu virus	 Antibodies amount in arbitrary unit (a.u.) <table border="1"> <thead> <tr> <th>Different antigens</th> <th>Antibodies amount (a.u.)</th> </tr> </thead> <tbody> <tr> <td>A</td> <td>~1</td> </tr> <tr> <td>B</td> <td>~1</td> </tr> <tr> <td>C</td> <td>~1</td> </tr> <tr> <td>D</td> <td>~1</td> </tr> </tbody> </table>	Different antigens	Antibodies amount (a.u.)	A	~1	B	~1	C	~1	D	~1
Different antigens	Antibodies amount (a.u.)											
A	~1											
B	~1											
C	~1											
D	~1											
 The same individual at the age of five years in contact with variant 2 of the flu virus	 Variant 2 of the flu virus	 Antibodies amount in arbitrary unit (a.u.) <table border="1"> <thead> <tr> <th>Different antigens</th> <th>Antibodies amount (a.u.)</th> </tr> </thead> <tbody> <tr> <td>A</td> <td>~5</td> </tr> <tr> <td>B</td> <td>~0.5</td> </tr> <tr> <td>C</td> <td>~5.5</td> </tr> <tr> <td>D</td> <td>~0.5</td> </tr> </tbody> </table>	Different antigens	Antibodies amount (a.u.)	A	~5	B	~0.5	C	~5.5	D	~0.5
Different antigens	Antibodies amount (a.u.)											
A	~5											
B	~0.5											
C	~5.5											
D	~0.5											
 The same individual at the age of 20 years in contact with variant 3 of the flu virus	 Variant 3 of the flu virus	 Antibodies amount in arbitrary unit (a.u.) <table border="1"> <thead> <tr> <th>Different antigens</th> <th>Antibodies amount (a.u.)</th> </tr> </thead> <tbody> <tr> <td>A</td> <td>~8</td> </tr> <tr> <td>B</td> <td>~0.5</td> </tr> <tr> <td>C</td> <td>~0.5</td> </tr> <tr> <td>D</td> <td>~5</td> </tr> </tbody> </table>	Different antigens	Antibodies amount (a.u.)	A	~8	B	~0.5	C	~0.5	D	~5
Different antigens	Antibodies amount (a.u.)											
A	~8											
B	~0.5											
C	~0.5											
D	~5											

- 1- Name the specific immune response revealed in the above document. Justify the answer.
- 2- Justify the following statements by referring to the document.
 - a- The secondary immune response is more amplified than the primary immune response.
 - b- The secreted antibody is specific to the antigen and not to the variant of the virus.
 - c- The organism keeps memory for an encountered antigen for more than ten years.
- 3- Name two cells implicated in the immune response triggered against variant 1 of the flu virus and specify the role of each cell.
- 4- Explain how the secreted antibodies contribute to the destruction of the flu virus.
- 5- Specify if the revealed immune response is capable alone to eliminate cells infected by the virus. Justify the answer.

Part of the Ex	Exercise 2 (5 points)	Mark
1	Specific humoral immune response; since following the entry of the virus of variant 1, having the antigens A, B, C and D, the amount of antibodies specific to each of these antigens rises to 1a.u..	1
2	<p>a- During the first contact with the variant 1 of flu virus at the age of 2 years, the amount of antibodies specific for each of the antigens A, B, C and D was 1a.u. which corresponds to a primary immune response . However, during the second contact at the age of 5 years with the variant 2 of flu virus having the antigens C and A in common with variant 1, the amounts of antibodies specific to A and to C increase respectively to 5 a.u. and to 6 a.u(> 1a.u) , while the amounts of antibodies specific to B and to D remains low (0.5 a.u.). This means that the immune response triggered upon the second contact with the same antigen or secondary immune response is more amplified than the primary immune response.</p> <p>b- During the 3rd contact at 20 years of age, with the variant 3 of flu virus having the antigens A and D in common with the variety 1, only the amount of antibody specific to these antigens increases respectively to 8 a.u (> 1a.u) and 6 a.u (> 1a.u) thus the secreted antibody is specific to the antigen and not to the variant of the virus.</p> <p>c- The organism keeps memory for an encountered antigen for more than ten years since the amount of antibodies has increased to 6 a.u(> 1a.u) 18 years after the first contact with the antigen D. Or Since the amount of anti B antibodies remains constant at 0.5 a.u from the age of 5 years to 20 years.</p>	2.25
3	<ul style="list-style-type: none"> - Macrophages: after phagocytosis of the antigen, they become APCs that activate specific T4L - T4L: once activated they secrete the interleukin 4 that activate the LB - LB: they identify the free antigens through their membrane antibodies to be activated - Plasma cells: secrete specific antibodies against the antigen. 	1
4	The specific antibodies neutralize their corresponding antigens of the flu virus by binding to them through their specific antigenic binding sites forming immune complexes. Thus the antibodies become able to bind through their constant part on macrophages that phagocytose the whole immune complexes thus destroying the virus (opsonization).	0.75
5	No. Since the infected cells cannot be identified by the antibodies which block only extracellular antigens.	0.5

الاسم : مسابقة في مادة "علوم الحياة"
الرقم : المدة ثلاثة ساعات

Answer the following exercises:

Exercise 1 (5 points)

The EBV virus infects 90% of the world population, but in a benign manner. This virus persists in the body. Its target cells are B lymphocytes.

Document 1 shows the activity of the EBV in “naive B Lymphocytes” (B lymphocytes that have never encountered the specific antigen) and in memory B lymphocytes specific for this antigen.

- Determine by referring to document 1, how the EBV virus persists and is produced in the body.

To better understand one of the immune responses triggered against the EBV virus, we follow up the evolution of anti-VCA and anti-EBNA antibodies directed respectively against two peptides VCA and EBNA that are found on the surface of this virus. The results are shown in document 2.

- Name the immune response revealed by these measurements. Justify the answer.
- Analyze the results of document 2. What can we draw out?

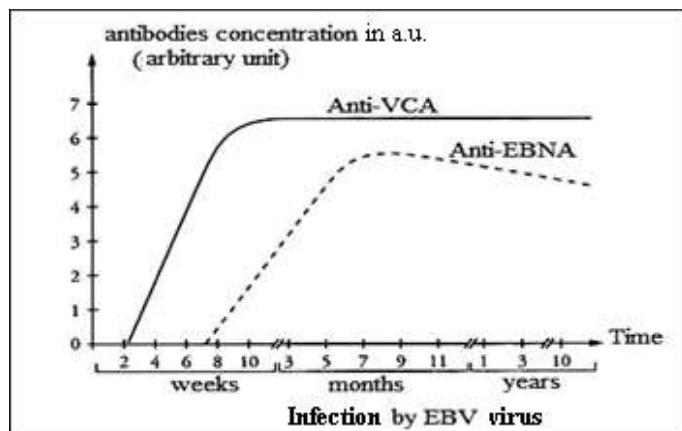
To Petri dishes containing appropriate culture medium, we add Lymphocytes (BL and TL) taken from different individuals infected or not by different viruses, EBV or other viruses. All the lymphocytes used in each experiment have the same HLA. Document 3 presents the conditions and the results of these experiments.

- Describe, in a short text, the experiments and the obtained results presented in document 3.
- Explain the obtained results of these experiments.

Immune responses against a virus

Activity of EBV	Naive B Lymphocyte	Memory B Lymphocyte
State of EBV in the lymphocyte	Active	Dormant
Presentation of viral peptides on the surface of the lymphocyte	Yes	No
Production of new viruses released into blood and able to infect other BL	Yes	No except if it is reactivated

Document 1



Document 2

Experiment	Experimental conditions	Results
1	T L of an individual infected by EBV ↓ BL infected by EBV	100% lysed BL
2	T L of an individual infected by EBV ↓ BL not infected by EBV	No lysed BL
3	T L of an individual infected by EBV ↓ memory BL infected by EBV	No lysed BL
4	T L of an individual infected by EBV ↓ BL infected by another virus	No lysed BL
5	T L of an individual not infected by EBV ↓ BL infected by EBV	No lysed BL

Document 3

Legend: → : Add

الاسم :

اسس التصحيح

الرقم :

مادة "علوم الحياة"

Part of the Ex	Answer key	Mark
	Exercise1 (5 points)	
1	<p>The virus persists in the body because it remains in the dormant state in the memory BL(0.25pt)</p> <p>The virus is produced by naive B lymphocytes that are once infected and by the memory BL once reactivated.(0.25pt)</p>	0.5
2	Specific humoral immune response (0.25pt) because the actors in this response are antibodies anti-VCA and anti- EBVA . (0.25pt)	0.5
3	<p>Anti-VCA antibodies appear in blood two weeks after infection and reach their maximum concentration 6.5 a. u. within eight weeks after the infection then stabilizes for the following 10 years. However Anti-EBVA antibodies appear later at the 7th week (7 w > 2 w) and reach their maximum concentration 5.5a.u. (5.5 < 6.5 a. u.) after more than 7 months (7months>8 w) then their concentration decreased slightly to reach 4.5 a. u. (4.5 < 6.5 a.u.) after 10 years. (1pt)</p> <p>This shows that the body develops two different humoral immune responses against two different peptides (different antigens) of EBV virus, and that the response triggered against the VCA is faster, more amplified and more sustainable than the one triggered against the EBNA. (0. 5pt)</p>	1.5
4	<p>Experiment 1: Lymphocytes TL of an individual infected with the virus EBV are added into the medium containing BL infected with EBV, we obtain 100% of lysis LB.</p> <p>Experiment 2: Lymphocytes TL of an individual infected with the virus EBV are added into the medium containing BL non infected with EBV, no lysis of BL is obtained.</p> <p>Experiment 3: Lymphocytes TL of an individual infected with the virus EBV are added into the medium containing memory BL infected with EBV, no lysis of BL is obtained.</p> <p>Experiment 4: Lymphocytes TL of an individual infected with the virus EBV are added into the medium containing BL infected with another virus, no lysis of BL is obtained.</p> <p>Experiment 5: Lymphocytes TL of an individual non infected with the virus EBV are added in the medium containing BL infected with EBV, no lysis of BL is obtained .</p>	1.25 (5 x0.25)
5	<p>The Lymphocytes T cytotoxic having receptors that recognize infected cells presenting at with their surface self HLA having non self peptide which has activated the same T8 lymphocytes which is identified in experiment 1 (there is 100% of lysed BL).</p> <p>In experiment 2, non-infected BL do not present non-self peptides this is why we do not observe any lysis.</p> <p>In experiment 3, B memory cells infected by the same virus as TL do not present non-self peptides. They are not identified by Tc and they are not lysed.</p> <p>In experience 4, BL infected by another virus present another non-self peptides. They are not identified by Tc and they are not lysed.</p> <p>In experiment 5, TL from an individual non-infected with EBV are not activated and differentiated into Tc and do not cause the lysis of BL infected by the virus.</p>	1.25 (5 x0.25)

Exercise 2 (5 points) Tetrahydrocannabinol and immune response

A recent experimental study was performed on mice to demonstrate the action of tetrahydrocannabinol (THC) on the immune system. THC is an active ingredient of cannabis (drug) that is suspected to modify the immune response against cancer cells. In order to study the tumor development and the immune response in the presence of THC, the following experiments were performed.

Experiment 1: We take two lots of non-immunized healthy mice, to which we implant cancer cells having the same strain as that of the mice: Lot 1 did not receive any injection of THC; Lot 2 was subjected to four injections of THC per week before and after the implantation of cancer cells. The results of the two lots are presented in documents 1 and 2.

Document 1 represents the variation of the tumor volume as a function of time after implantation. Document 2 shows the proliferation of T lymphocytes as a function of the percentage of the cancer cells implanted relative to the number of T lymphocytes before proliferation.

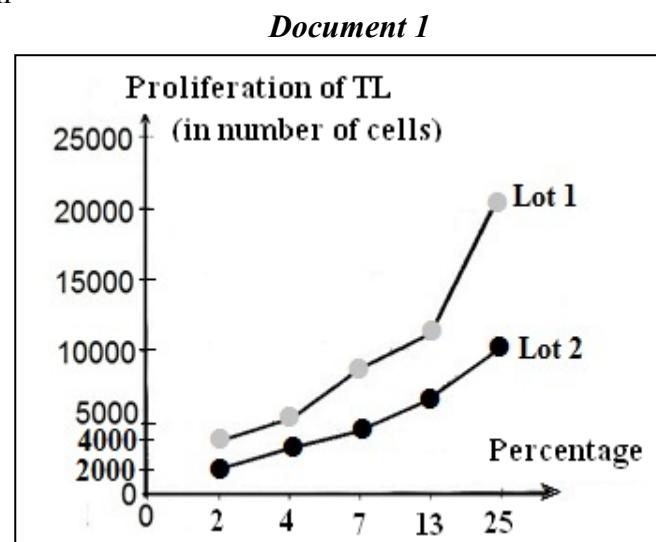
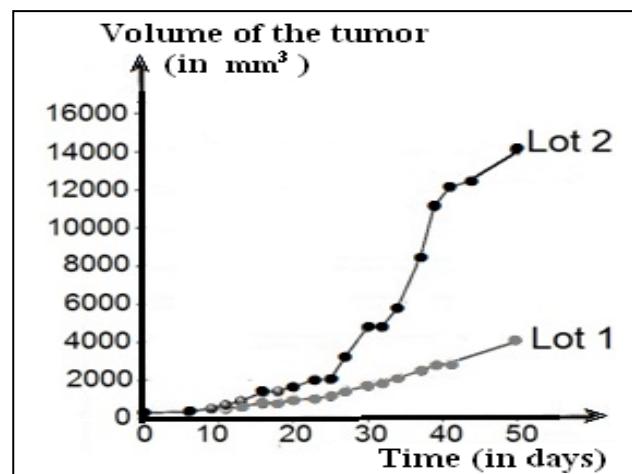
- Interpret the results of each of the documents 1 and 2.

Experiment 2: The level of interleukins secreted in the mice of experiment 1 was measured at the level of the tumor and the spleen. These values are presented in document 3.

- Determine, with reference to document 3 and to acquired knowledge, the target cells of THC.
- Draw out referring to all what preceded, the action of THC on the immune response against the tumor.

In the framework of studying the effect of THC on the secondary immune response, we perform experiment 3.

Experiment 3: New mice belonging to the same lots are immunized against this tumor before being subjected to implantation of cancer cells. From each lot, eight mice were subjected to the implantation of a variable number of cancer cells and the percentage of mice rejecting the tumor was calculated (Document 4).



	Interleukins secreted at the level of the tumor (pg.mL ⁻¹ for 500 mg of tumor)	Interleukins secreted at the level of the spleen (pg.mL ⁻¹ for 10 ⁶ cells)
Lot 1	190	37
Lot 2	73	21

Document 3

Number of live implanted tumor cells	1 x 10 ⁵			2 x 10 ⁵			3 x 10 ⁵		
	1 x 10 ⁵	2 x 10 ⁵	3 x 10 ⁵	1 x 10 ⁵	2 x 10 ⁵	3 x 10 ⁵	1 x 10 ⁵	2 x 10 ⁵	3 x 10 ⁵
Percentage of mice rejecting the tumor	Lot 1	100%	100%	100%	100%	100%	60%	60%	50%
	Lot 2	100%	100%	100%	100%	100%	60%	60%	50%

- Construct a histogram that translates the results of document 4.
- Analyze the results of experiment 3. What can you draw out?

Document 4

Part of ex.	Answer	Grade												
	Exercise 2													
1	<p>Document 1: The volume of the tumor (VT) is constant between day 0 and day 10 in both lots. On the other hand, this volume increases to 4000mm³ between day 10 and day 50 in lot 1. Similarly, in lot 2 which received THC injection VT increases slightly to 2000 mm³ from day 10 till day 25 followed by a sharp increase to a much higher value than lot 1 (14000 mm³) between day 25 and day 50. This shows that THC favors the development of the tumor after 10 days(1/2 pt)</p> <p>Document 2: The number of T cells in the mice of lot 1 increases from 4000 cells to 20000 cells when the percentage of implanted tumor cells relative to the number of lymphocytes before proliferation increases from 2 % to 25%. Similarly, in lot 2 which received THC, the number of TL increases but slightly from 2000(< 4000) to 10000(< 20000).</p> <p>This shows that the proliferation of TL varies in the same direction as the quantity of cancer cells and THC reduces this rate of proliferation against tumor cells. (1/2 pt)</p>	1												
2	The level of IL secreted at the level of the tumor in lot 2 is 73 pg / mL for 500 mg of tumor which is less than 190 pg / mL for Lot 1. Similarly, in lot 2 the level of IL secreted at the level of the spleen is 21 pg / mL for 10 ⁶ cells which is less than that secreted at the level of the spleen of Lot 1 (37 pg / mL for 10 ⁶ cells). Thus THC decreases the secretion of interleukins and since interleukins are secreted by T4 cells then the target cells of THC are the T4cells.	1												
3	THC acts on T4 lymphocytes secreting IL indispensable for the activation of the specific immune response (humoral mediated and cell mediated). Thus a decrease in the quantity of IL provokes a decrease in the proliferation of T lymphocytes leading to a decrease in the immune response. Hence the tumor develops.	3/4												
4	<p>Histogram: percentage of mice rejecting the tumor as a function of number of live implanted tumor cells.</p> <p>Percentage of mice rejecting the tumor</p> <table border="1"> <caption>Data extracted from the histogram</caption> <thead> <tr> <th>Number of live implanted tumor cells</th> <th>Lot 1 (%)</th> <th>Lot 2 (%)</th> </tr> </thead> <tbody> <tr> <td>1x10⁵</td> <td>100</td> <td>100</td> </tr> <tr> <td>2x10⁵</td> <td>100</td> <td>~60</td> </tr> <tr> <td>3x10⁵</td> <td>100</td> <td>~50</td> </tr> </tbody> </table> <p>Scale □ : 20%</p> <p>Legend ■ : Lot 1 □ : Lot 2</p> <p>Number of live implanted tumor cells</p>	Number of live implanted tumor cells	Lot 1 (%)	Lot 2 (%)	1x10 ⁵	100	100	2x10 ⁵	100	~60	3x10 ⁵	100	~50	1 1/2
Number of live implanted tumor cells	Lot 1 (%)	Lot 2 (%)												
1x10 ⁵	100	100												
2x10 ⁵	100	~60												
3x10 ⁵	100	~50												
5	<p>Document 4 shows that the percentage of mice rejecting the tumor in lot 1 is constant 100 percent regardless of the number of implanted tumor cells while in lot 2 receiving injections of THC, it decreases from 100 % to 50% when the number of implanted tumor cells increases from 1x10⁵ to 3x10⁵. (1/2)</p> <p>In mice immunized against this tumor, the rejection of the tumor corresponds to a secondary immune response. Hence THC weakens the secondary immune response. (1/4pt)</p>	3/4												

Exercise 2 (5 points)**Cellular Cooperation and Production of Antibodies**

In the framework of determining the conditions of the production of antibodies during the immune response, we perform a series of experiments on mice of the same strain.

Experiment 1: Mice are subjected to the ablation of the thymus followed by irradiation that destroys all cells of the immune system. These mice are then divided into 4 lots and treated as shown in document 1.

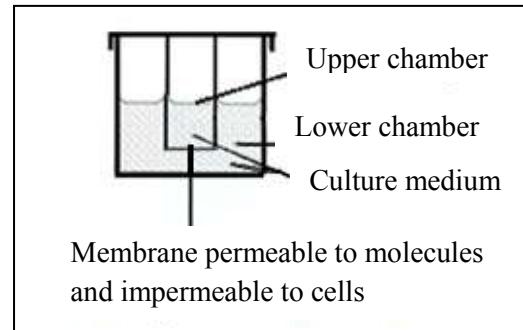
Ablation of the thymus then irradiation of the mice				
	Lot 1	Lot 2	Lot 3	Lot 4
Injection of lymphocytes removed from mice of the same strain	T	B and T	B and T	B
Injection of an antigen: SRBC (sheep red blood cells)	Yes	Yes	No	Yes
One week later, removal of serum from the mice and addition of SRBC to the serum				
Results : agglutination of SRBC	absence	presence	absence	absence

Document 1

- Interpret the experimental results of experiment 1.
- Specify the aim of destroying the cells of the immune system before starting the experiment.

Experiment 2: A mouse receives an injection of sheep red blood cells (SRBC). Three days later, we extract lymphocytes from its spleen. These lymphocytes are distributed into 4 identical lots then cultured in Marbrook chamber (document 2) according to the procedure described in document 3.

Few days later, the culture medium is filtered and the collected liquid is added to SRBC. The results are shown in document 3.

**Document 2**

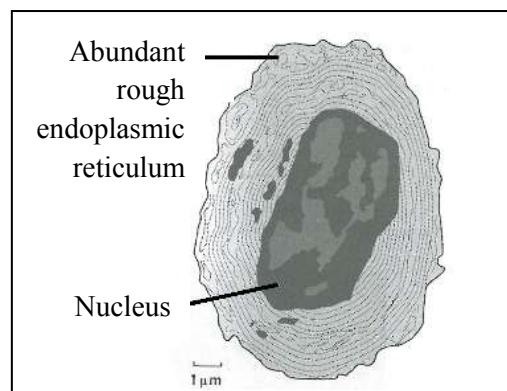
Culture medium	1	2	3	4
Lymphocytes placed in the upper chamber	none	T	none	none
Lymphocytes placed in the lower chamber	T and B	B	B	T
Results : agglutination of SRBC	Strong	Strong	Null	Null

Document 3

- Analyze the results of media 1 and 2. What can you draw out?

Document 4 illustrates an electronography of an antibody secreting cell that is found in large quantities, in media 1 and 2 of document 3 and absent in media 3 and 4.

- Name this cell. Justify the answer.
- Explain the variation in the quantity of this type of cells in the four media of experiment 2.

**Document 4**

Part of ex	Answer key	Note
	Exercise 2 (5 points)	
1	<p>There is agglutination of SRBC in lot 2 where there was an injection of lymphocytes B and T and SRBC at the same time whereas there is no agglutination in lot 1 where there were only injection of lymphocytes T with SRBC and in lot 4 where there are only injection of B lymphocytes with SRBC; This shows that the agglutination requires the cooperation of TL and BL or the presence of the TL and BL at the same time.</p>	1 1/2
2	<p>There is agglutination of SRBC in lot 2 where there was an injection of lymphocytes B and T and SRBC at the same time however there is no agglutination in lot 3 where there were injection of lymphocytes B and T without SRBC. This shows that the contact with the antigen a week in advance is necessary to get agglutination.</p>	1/2
3	<p>There is strong agglutination of SRBC in the media 1 and 2 where B and T lymphocytes are found together whether they are in the same medium (medium 1) or separated by a membrane that is impermeable to cells but permeable to molecules (medium 2). (1/2 pt) Therefore, the agglutination of SRBC that is due to the production of anti-SRBC antibodies requires the cooperation of B and T lymphocytes via molecules and not by direct contact. (1/2 pt)</p>	1
4	<p>Plasmocyte (1/2 pt) because this cell has a voluminous cytoplasm that is rich in rough endoplasmic reticulum, cytoplasmic organelle that is indispensable for the synthesis of proteins such as antibodies. (1/2 pt)</p>	1
5	<p>Plasmocytes are derived from the differentiation of lymphocytes B which are absent in medium 4 where there is only TL, hence plasmocytes are absent in this medium. The differentiation of LB into plasmocytes is stimulated by IL 4 that is secreted by TL that are absent in medium 3, hence plasmocytes are absent in this medium. However B and T cells are present in media 1 and 2. IL 4 stimulates directly the B cells in the lower chamber (medium 1) or crosses the permeable membrane and stimulates B cells (medium 2). Hence the abundance of plasmocytes in these two media.</p>	1

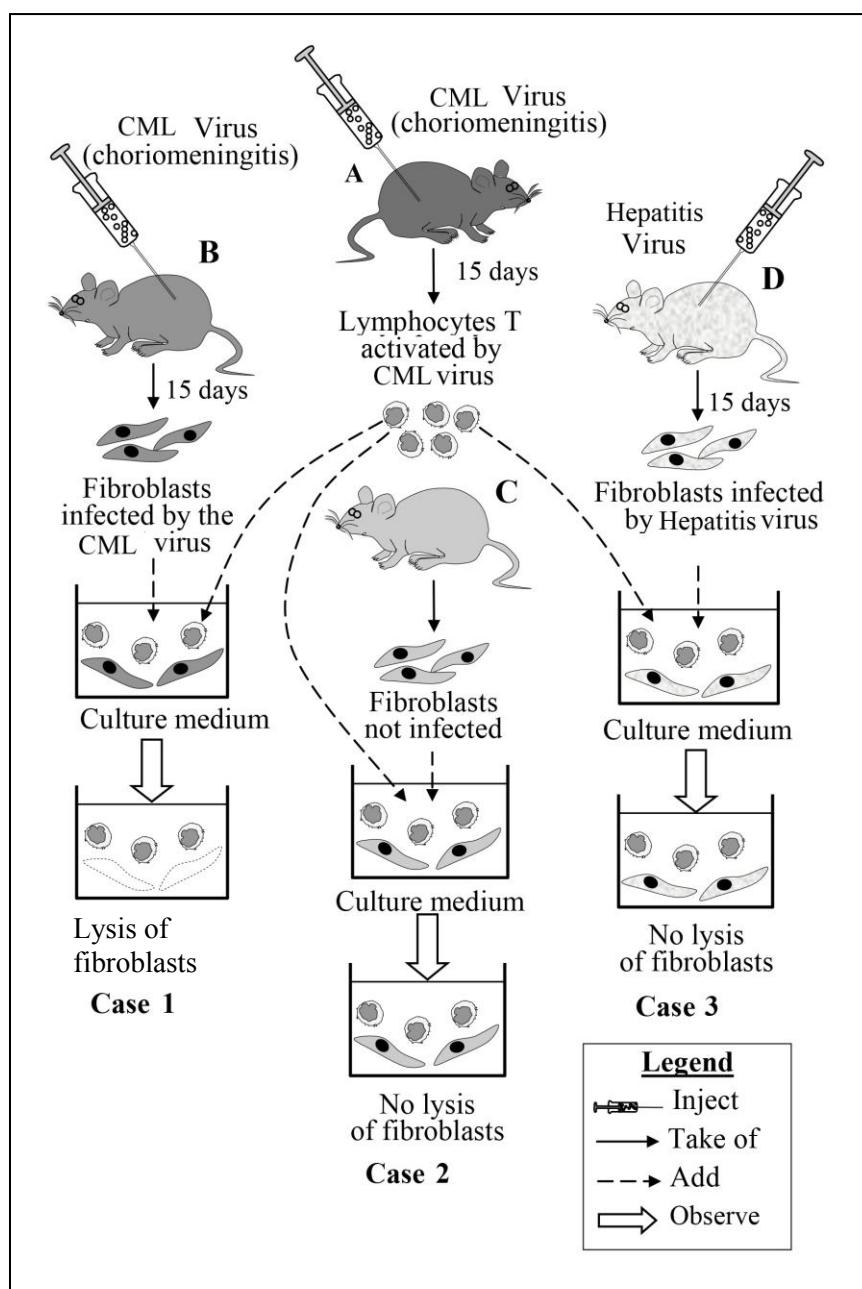
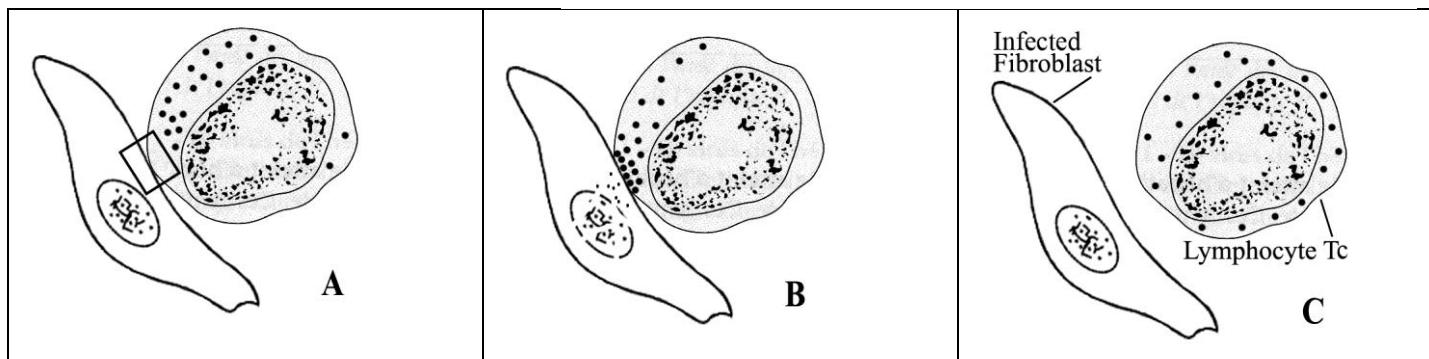
Exercise 2 (5 points)

Choriomeningitis virus (CML) is a virus transmitted by rodents. The disease is manifested by symptoms similar to those of flu with fever. This disease is transmitted to humans by contaminated food or dust from infected mice.

To better understand the immune mechanisms responsible for the lysis of infected cells, a set of experiments are performed on mice of the same line (document1).

1. Pick out from the text the means of contamination of humans by CML virus.
2. Describe the experiments schematized in document 1.
3. Interpret the results of the experiments of document 1.

Document 2 shows the schematic representations of the cellular interactions observed in the culture medium in case1.

**Document 1****Document 2**

4. Arrange, in chronological order, the schematic representations of document 2. Justify the answer.
5. Explain the mechanism of cell lysis observed in document 2.

Part	Answer key Exercise 2	Grade
1	The means of human contamination by the CML virus are: food and dust contaminated by infected mice.	0,5
2	Mouse A is injected with CML virus (choriomeningitis). 15 days later T lymphocytes activated by CML of this mouse are taken of and added to three culture media. Mouse B is injected with CML virus (choriomeningitis). 15 days later, the fibroblasts infected by CML of this mouse are taken of and are added with the activated T lymphocytes of mouse A to a culture medium. Lysis of these fibroblasts is observed. Non infected fibroblasts of mouse C are taken of and added with the activated T lymphocytes of mouse A to a culture medium. No lysis of these fibroblasts is observed. Mouse D is injected with hepatitis virus. 15 days later, the fibroblasts infected by hepatitis virus of this mouse are added with activated T lymphocytes of mouse A to a culture medium. No lysis of these fibroblasts is observed.	1,5
3	There is lysis of the fibroblasts of mouse B that are infected by the CML virus in the medium containing T lymphocytes activated by the same virus, while there's no lysis of non-infected fibroblasts of the mouse C neither of the fibroblasts of the mouse D that are infected by another virus (hepatitis virus) which are placed in a culture medium containing the same T lymphocytes. This shows that activated T lymphocytes destroy only the cells that are infected by the same virus that led to their activation OR activated T lymphocytes destroy only the cells that are infected and that they are specific to the CML antigen.	1
4	1- The order is: C A B (0,25) The first scheme C shows near the infected fibroblast one T lymphocyte with vesicles that are spread in its cytoplasm. In the second scheme A, the T lymphocyte is in contact with the membrane of the infected fibroblast, what corresponds to the double recognition. In the third scheme B, we notice that the granules are in contact with the infected fibroblast and destroy its nucleus. (0,75 pt)	1
5	Tc recognizes the infected body cell and binds by its TCR to the self HLA-I non self peptide complex expressed on the membrane of the infected cell. Then it liberates perforin to form polyperforin channels through the membrane of the infected cell. After that the TcL releases granzymes that penetrates into the infected cell through the polyperforin channels leading to the degradation of its DNA, thus causing lysis of the infected cell.	1

Exercise 2 (5 points)**Graft Rejection**

The early grafts done on humans failed: they undergo necrosis (important destruction of the graft cells) leading sometimes to death.

In order to understand how the rejection of the graft takes place, several experiments and studies are performed.

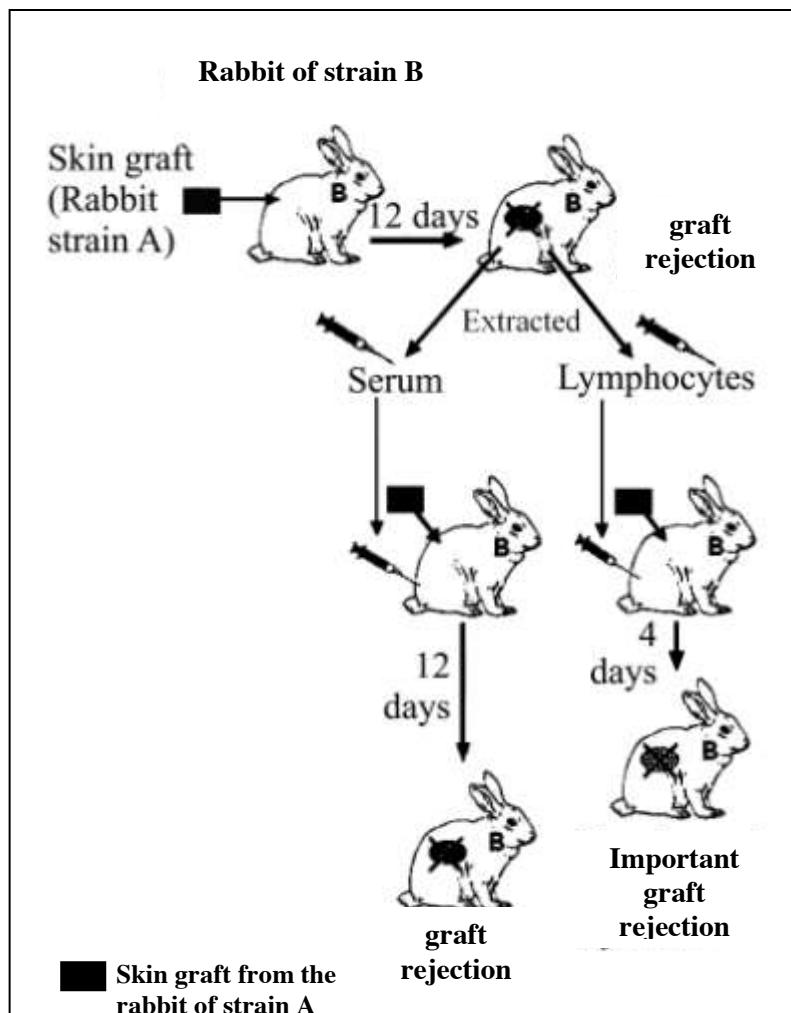
When skin grafts are done between two individuals that are genetically different, first the graft is richly vascularized; then, within 7 to 14 days, it is invaded by white blood cells and undergoes necrosis. This is graft rejection.

- 1-** Draw out from the text:

- 1.1-** One essential condition for the acceptance of the graft.
- 1.2-** The duration necessary for graft rejection.

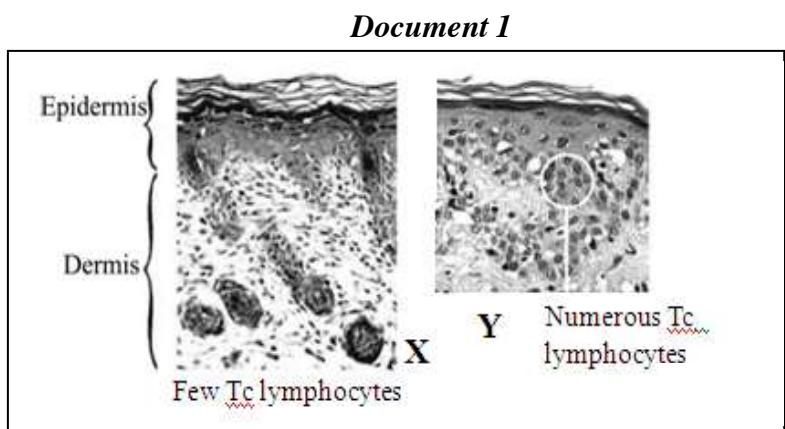
Peter Brian Medawar performed a skin graft experiment on rabbits A and B of different strains (genetically different). This experiment is presented in document 1.

- 2-** Interpret the results of this experiment.
What can you conclude?



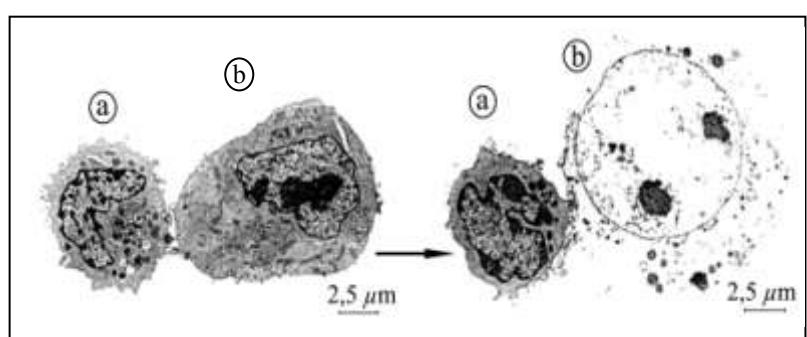
Document 2 shows histological sections of the skin of a mouse:

- X: normal skin
 - Y: fragment of a skin which is grafted in a mouse of different strain and observed five days after grafting.
- 3-** Compare the two histological sections of skin, X and Y.
- Draw out the type of the immune response triggered against the graft.



Document 3 shows the electronographs of two mouse cells which are cultured in vitro: a cytotoxic T lymphocyte (a) and a target cell (b). These electronographs are observed at two different moments during graft rejection.

- 4-** Name the mechanism shown in document 3.
- 5-** Explain briefly this mechanism.

**Document 3**

Part	Answer key	Mark
Exercise 2		
1	<p>1.1 The graft is accepted when done between two genetically identical individuals.</p> <p>1.2 Between 7 and 14 days (12 days).</p>	0.5
2	<p>The skin graft A in rabbit of strain B, is always rejected within 12 days. Similarly, it is rejected, within 12 days; by a rabbit B of strain B even if it has received serum from another rabbit of strain B who has already rejected the graft. This shows that, the serum has no effect on the response of the graft rejection or it does not contain the effectors of the immune response.</p> <p>On the contrary, it will be rejected rapidly, in 4 days, if the rabbit of strain B receives the lymphocytes taken from a rabbit of strain B that has already rejected the graft from A. This shows that the lymphocytes have accelerated the graft rejection and are the effectors of the response triggered against the graft.</p> <p>We conclude that lymphocytes are the effectors of the immune response triggered against graft between individual of different strains.</p>	1.5
3	<p>The histological section of the normal skin shows a thicker dermis (12mm) compared to that of the skin that has received the graft (8 mm).</p> <p>The histological section of normal skin shows fewer Tc lymphocytes compared to that of the skin that has received the graft. The histological section of normal skin shows few Tc lymphocytes while they are more abundant in the skin that has received the graft. Both have epidermis of the same thickness.</p> <p>It is a cell mediated immune response.</p>	1.5
4	Cell Lysis (apoptosis)	0.5
5	<p>Tc recognizes the infected body cell and binds through its TCR to MHC I – non self peptide complex on the membrane of the infected cell. Then it liberates perforin to form polyperforin channels through the membrane of the infected cell. After that, the Tc cell releases granzymes into the infected cell through the polyperforin channels leading to the degradation of its DNA, thus causing lysis of the infected cell.</p>	1

الاسم: مسابقة في مادة علوم الحياة
الرقم: المدة: ثلاثة ساعات

Answer the following exercises

Exercise 1 (5 points)

Vaccine against AIDS

In the framework of researches concerning AIDS, scientists followed up 1600 non treated persons that are infected by HIV (Human Immunodeficiency Virus). They measured, at the beginning of the infection, the viral concentration in the blood and recorded the percentage of persons reaching the phase of AIDS. The results are presented in document 1.

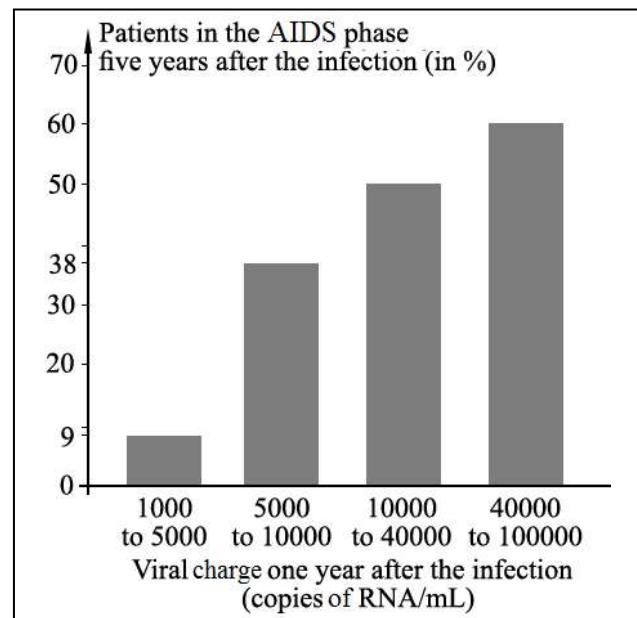
- Justify, by referring to document 1, the following statement: "in the absence of treatment, there is a relation between the onset of the phase of AIDS and the early evolution of the viral charge".

In the case of HIV, vaccines that activate only the production of anti-HIV antibodies don't protect against all the known strains of the virus. Currently, the scientific community agrees on the fact that: to be effective, a vaccine should also stimulate the production of cytotoxic T lymphocytes directed against HIV. This allowed the elaboration of vaccines against HIV.

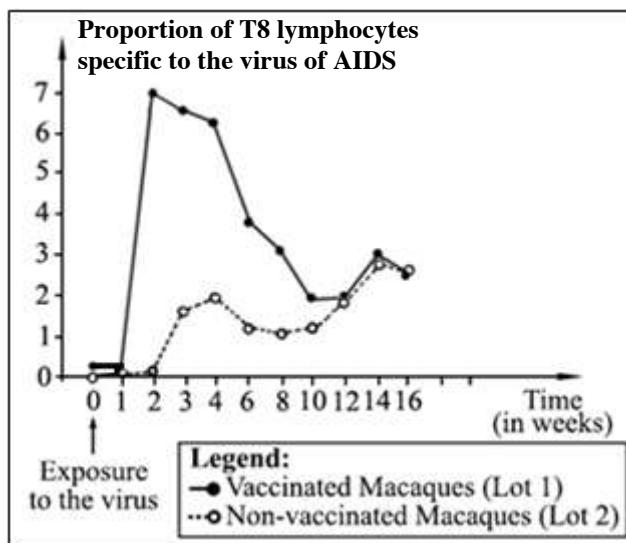
- Indicate how vaccination protects against a given antigen.

One of these vaccines was tested on two lots of macaques monkeys that are not infected by the virus of AIDS. The monkeys of the first lot (lot1) receives a series of five vaccine injections. The monkeys of the second lot (lot 2) are not vaccinated. Then, all the monkeys are exposed to the virus. The proportion of T8 lymphocytes specific to the AIDS virus is then evaluated in the blood of the monkeys (document 2). The viral charge is measured in the two lots of monkeys at the 8th and at the 24th week following the exposure to the virus (document 3).

- Show, by referring to document 2, that the immune response of the vaccinated monkeys is more rapid and more amplified than that of the non-vaccinated monkeys during the first 3 months of the infection.
- Determine if the immune response triggered in lot 1 is durable.
- Interpret the results of document 3.
- Show, by referring to what precedes, that the tested vaccine has a limited efficiency and doesn't allow the eradication of the disease.



Document 1



Document 2

Time after exposure to the virus.	Viral charge (number of viral RNA copies / ml of plasma)	
	Lot 1	Lot 2
8 th week	5.10^4	25.10^4
24 th week	5.10^4	50.10^4

Document 3

Part	Answer Exercise 1	Grade
1	The percentage of untreated patients in the AIDS phase five years after the infection increases from 9% to 60% when the viral charge one year after the infection increases from 1000 up to 10000 copies of mRNA/mL. This shows that the onset of the AIDS phase and the early evolution of the viral charge vary in parallel to each other.	1
2	Vaccines immunize the organism against a specific antigen by inducing a durable immunological memory. Or Vaccines protect the organism by sensitizing the immune system against the pathogens in order to recognize and destroy them in a more rapid and more efficient manner upon a second contact with the same pathogen.	0,5
3	In vaccinated monkeys, the proportion of T8 lymphocytes increases from 0 to 6.5 greater than that of non-vaccinated monkeys which is 2. This shows that the response triggered in vaccinated monkeys is more amplified. (0,5 pt) After the exposure to the virus, the increase of the proportion of T8 lymphocytes in vaccinated monkey begins after a latency time of one week, less than that in the vaccinated monkeys which is 2 weeks. This shows that the response in vaccinated monkeys is more rapid than that of the non-vaccinated ones. (0,5 pt)	1
4	Between the fourth and the 12 th week, the proportion of T8 lymphocytes, in both lots 1 and 2 decreases while remaining higher in vaccinated monkeys and reach the same value of 2 at week 12. After the 12 th week, the variations of these proportions remain identical. This shows that the immune response triggered by the vaccine is not durable, it does not last except for 12 weeks.	0,5
5	Document 3 shows that the viral charge at the 8th week in vaccinated macaques is $5 \cdot 10^4$ viral RNA copies/ml inferior to that of the non-vaccinated ones $25 \cdot 10^4$ viral RNA copies/ml . At the 24 th week it increases (doubles) to $50 \cdot 10^4$ viral RNA copies/ml in the non-vaccinated macaques while it remains constant at $5 \cdot 10^4$ viral RNA copies/ml in vaccinated macaques value that is 10 times smaller than $50 \cdot 10^4$ viral RNA copies/ml. This shows that vaccine maintains the viral charge weak and constant at the beginning of the infection.	1
6	The chance of reaching the phase of AIDS diminishes in the case where the viral charge is weak at the beginning of the infection (doc.1). The vaccine maintains the viral charge low at the beginning of the infection (doc.3). This diminishes the evolutions of the disease toward the phase of AIDS thus extending the asymptomatic phase. Therefore there is a greater chance to prolong the life of seropositive individuals. From this point the vaccine is efficient. The vaccine amplifies the specific cell mediated immune response the first three months after infection (doc.2) however this amplification is not durable thus the efficiency is limited. In addition the vaccine doesn't ensure a total recovery and the disease is not eradicated.	1

Infection by HIV

Exercise 2 (5 points)

HIV is a retrovirus whose genetic material consists of RNA. When the virus infects a target cell, it uses the transcription and translation machineries of the host cell in order to synthesize its different components. The infected cell produces new viruses and then dies.

- 1- Indicate the type of the specific immune response that is triggered against cells infected by a virus. Justify the answer.

To better understand the mechanism of infection of a target cell by HIV, the following studies are performed:

Leucocytes are cultured then exposed to HIV. The survival of these cells is measured during the days that follow the exposure. The results are presented in document 1.

- 2- Represent in a table the obtained results shown in document 1.
- 3- 3-1 Analyze the obtained results (doc 1).
3-2 Conclude the target cells of HIV.

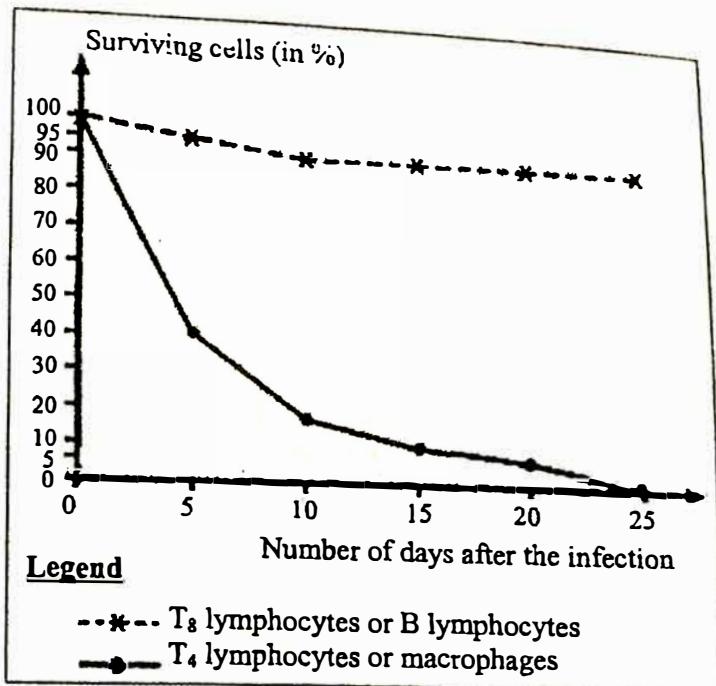
Document 2 shows the stage of recognition of a target cell by the HIV.

- 4- Specify, by referring to document 2, the characteristic that allows a cell to be a target for HIV.

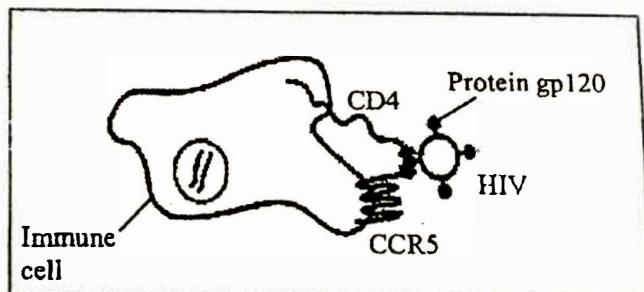
In order to identify the infected cell, two immune cells 1 and 2 are extracted from a patient and isolated. DNA probe having complementary sequence to the HIV viral genome is added. Once fixed, the probe is visualized by fluorescence using an appropriate technique.

The obtained results are presented in document 3.
For simplification, only three pairs of chromosomes are represented.

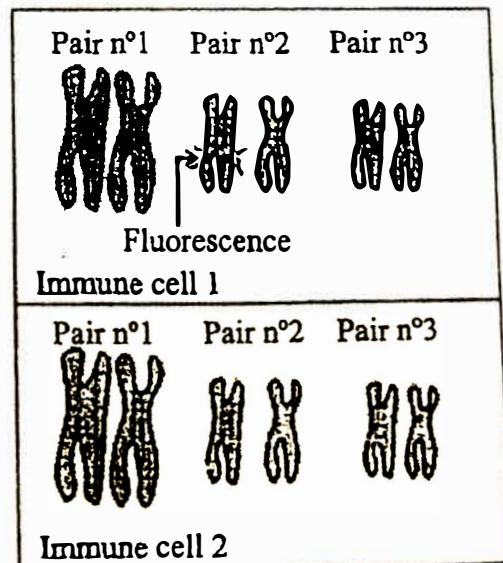
- 5- Identify which immune cell (1 or 2) is infected by HIV.
- 6- Explain the mode of infection of an immune cell by HIV starting from its recognition till the production of new viruses.



Document 1



Document 2



Document 3

Days after infection	0	5	10	15	20	25
T ₈ Lymphocyte or B lymphocytes%	100	95	90	90	90	90
T ₄ lymphocytes or macrophage%	100	40	18	10	5	0

2) Title: the variation in the percentage of the surviving leucocytes cells as a function of days after infection with HIV.

- 3.1- The number of surviving cells T₈ lymphocytes or B lymphocytes decrease very slightly during the first 10 days from 100% to 95% after the infection (exposing to HIV), then remains constant in the next 15 days, while the number of surviving cells T₄ lymphocytes or macrophage decreases sharply from 100% to 20% in 10 days, then continues to decrease to become zero at day 25.

- 3.2- The target cells of HIV are T₄ lymphocytes or macrophage.

4) The presence of receptors called CD₄ and co receptor CCR₅ at the level of cell surface, since from doc.2 the protein gp120 of the HIV has the capacity to recognize and bind to an immune cell having these two receptors.

5) The DNA probe binds to HIV viral genome is visualized by fluorescence which is observed in the chromosome pair number 2 in the immune cell 1 and not in the immune cell 2. This means that the immune cell 1 is infected by HIV.

6) The protein gp120 of HIV recognizes and binds with receptors CD₄ and CCR₅ presented at the surface of the immune cell T₄ or macrophage, leading to the entrance of the viral genetic material RNA into the host cell in which it changes into the viral DNA under the effect of reverse transcriptase to bind with DNA of the host cell. Then this viral DNA undergoes transcription in the nucleus of the host cell, finally the RNA of virus undergoes translation in the ribosome in the cytoplasm leading to the production of different viral components needed to produce new viruses.

Exercise 2

- 1) Cell mediated immune response, since the virus infects cells which result in its death, meaning that the body triggers an immune response against infected cells through specific immune cell (T_c)

Exercise 4 (5 points)**Fight Against Ebola**

Ebola is a very contagious and fatal virus that causes hemorrhagic fever. It is transmitted through blood, saliva, feces as well as through sexual contacts.

Infected individuals who survived, show first a high amount of specific anti-Ebola antibodies, followed by the disappearance of the virus with an important increase in specific cytotoxic T cells (TcL).

- 1-** Identify the immune response(s) triggered against Ebola.

In order to develop fighting or therapeutic modalities against this disease, researchers performed experiments that are described below.

- In December 2011, researchers developed a vaccine. They isolated a surface protein of the virus and injected it to a first lot of mice. To a second lot, they injected the same protein in the form of immune complexes called EIC (Ebola Immune Complexes). To a third lot they injected the EIC and a substance, the PIC. The injections are repeated four times for each lot. Two weeks after each injection, serum is collected from the mice and the antibodies amounts were measured. The obtained results are presented in document 1.

- 2-** Determine the most efficient vaccine against Ebola.

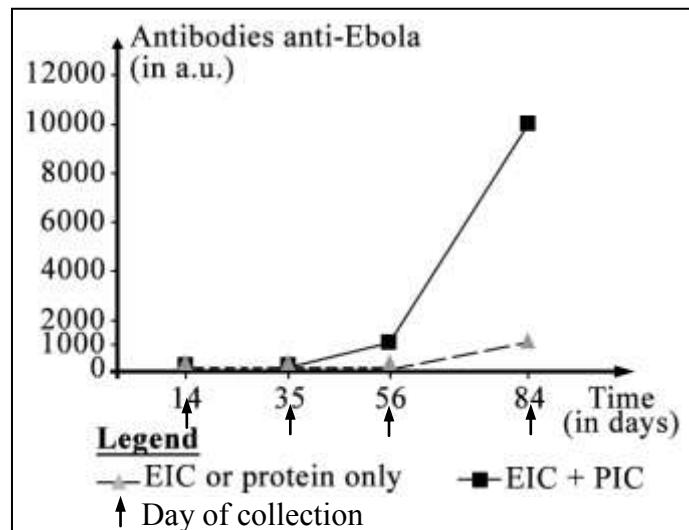
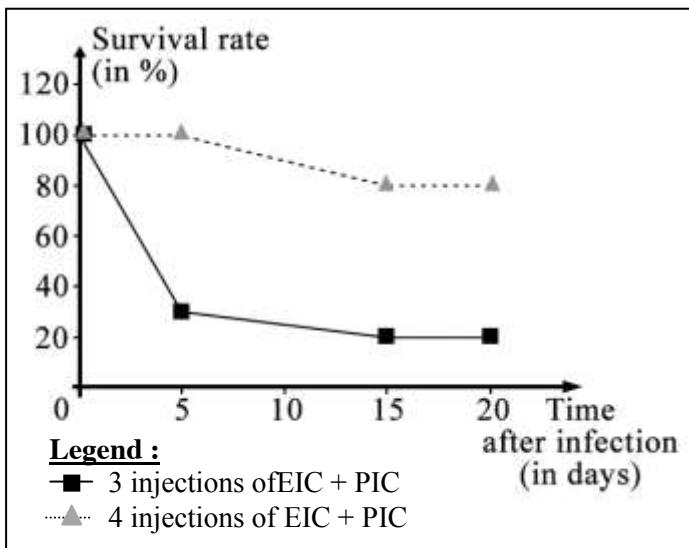
The molecule PIC is an agonist to proteins that are indispensable for phagocytosis.

- 3-** Indicate the roles and the moments where macrophages intervene in the specific immune response triggered against Ebola.

Two lots of mice have been vaccinated using the mixture EIC+PIC, the first lot received three boosters for the vaccine and the second received four boosters. After that both lots were contaminated by Ebola virus. The results concerning the survival of the mice are presented in document 2.

- 4-** Deduce one condition for the vaccination against Ebola to be successful.

- In June 2012, Canadian researchers performed the following experiment: two lots of monkeys, infected by the Ebola virus, received a mixture of three antibodies specific to particular epitopes of the virus. The obtained results are presented in document 3.

**Document 1****Document 2**

Lots of monkeys	Performed treatment	Number of monkeys	Number of surviving monkeys
A	Infection by the virus then injection of antibodies 24 hours after infection	4	4
B	Infection by the virus then injection of antibodies 48 hours after infection	4	2

Document 3

- 5-** Explain the obtained results.

- 6-** Distinguish serotherapy from vaccination concerning: the nature of the injected substance, the latency period and the duration of the protection established against Ebola.

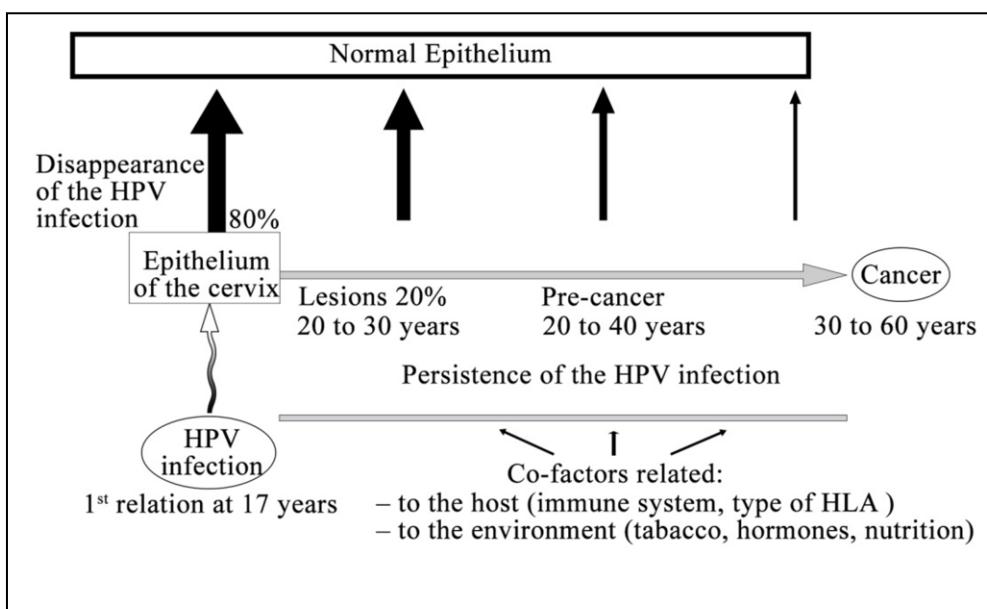
Part of the exercise	Exercise4 Fight against Ebola	Grade 5 pts
1	<p>A humoral specific immune response is triggered, since in case of Ebola the surviving individuals have a high amount of anti-Ebola antibodies that are released by plasma cells that are the effectors of humoral specific immune response.</p> <p>A cell-mediated specific immune response is triggered, since the surviving individuals show an important increase in the specific TcL which are the effectors of the cell mediated specific immune response.</p>	$\frac{1}{2}$ $\frac{1}{2}$
2	<p>The amount of anti-Ebola antibodies is nil and remains constant on the 14th and 35th day, after the first and the second injection of the three types of vaccine. After the 3rd injection of vaccine, this amount increases to 1000 a.u on the 56th day in individuals having received EIC + PIC, while it remains constant and nil in individuals having received the vaccine EIC or only the protein. This amount of antibodies increases in the three lots to reach 10000 a.u on the 84th day in individuals having received the vaccine EIC+PIC which is 10 times higher than the 1000a.u obtained when only the vaccine EIC or only the protein is administered. This shows that the vaccine EIC+PIC is the most effective.</p>	3/4
3	<p>At the beginning of the specific immune response, macrophages act as antigen presenting cells which induce the specific immune response.</p> <p>At the end of the specific humoral immune response, they perform phagocytosis of the immune complex in order to eliminate antigens.</p>	1
4	<p>Between day 0 and day 20, the percentage of survival decreases from 100% to 80% in the lot receiving 4 injections. This decrease is 4 times more significant than that obtained in the lot receiving only 3 injections which reaches 20 %. Thus, the condition for the vaccination against Ebola to be successful is to give 4 boosters.</p>	1/2
5	<p>The antibodies injected after 24 hours neutralize the antigen and slow down efficiently the propagation of virus which allows the body defenses to react and protect all the monkeys (4/4) which remain alive. However, when the injection is delayed to 48 hours, the viruses multiply more rapidly than the lymphocytes involved in the specific immune response and infect a great number of cells before being neutralized by the specific injected antibodies. This reduces the efficiency of the body defense and sometimes renders it insufficient. This explains the death of two out of the four infected monkeys.</p>	1
6	<p>In serotherapy, the injected substances are the specific antibodies while in vaccination, the injected substances are viral or antigenic proteins.</p> <p>In serotherapy, the latency time is null while in vaccination, the latency time is 2 weeks</p> <p>In serotherapy, the duration of protection time is short while in vaccination, the protection is more durable.</p>	$\frac{1}{4}$ $\frac{1}{4}$ $\frac{1}{4}$

Exercise 2 (5 points)

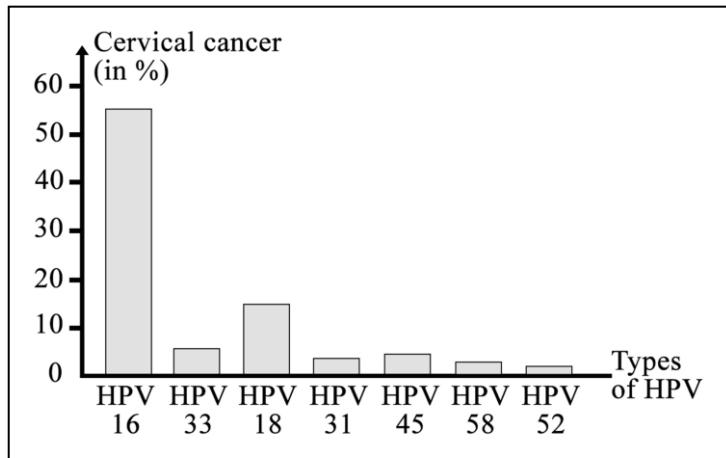
Cervical cancer is considered to be the second cancer that affects women in the developing countries, and the eighth in the developed ones. To better understand the cause of this cancer and in order to prevent it efficiently, researchers performed different studies.

- A study involving thousands of women suffering from cervical cancer shows that 75% of these women have encountered the human papillomavirus (HPV) during their sexual life.

Document 1 shows the evolution of the state of the cervical epithelium after HPV infection.

**Document 1**

- 1- Justify the following statement: « Cervical cancer is a slowly induced viral cancer ».
- 2- Pick out, from document 1, two other risk factors for the development of cervical cancer.
- 3- Indicate the type of specific immune response triggered to fight against a viral infection. Justify the answer.
- In a second study, 150 types of HPV have been identified, some of which are qualified as being of "high-risk", induce a genetic mutation which causes cervical cancer. Document 2 shows the percentages of women having cervical cancer as a function of the types of HPV that have infected them.
- 4- Deduce from document 2 the two types of HPV of high risk .

**Document 2**

- Researchers have elaborated prophylactic vaccines aiming to ensure a preventive protection of individuals against the infection. These vaccines stimulate the production of antibodies directed against certain types of HPV viruses. The characteristics of two of these vaccines are regrouped in document 3.

- 5- Determine the most efficient vaccine.
- 6- Explain how the antibodies produced during this vaccination allow protection against cervical cancer.
- 7- Suggest two preventive means against cervical cancer.

	Vaccine	
	Gardasil	Cervarix
Types of targeted HPV	Quadrivalent Vaccine HPV6, HPV11, HPV16 and HPV18	Bivalent Vaccine HPV16 and HPV18
Suggested amount	Almost 20 µg	20 µg
Vaccination Schedule	0, 2 and 6 months	0, 1 and 6 months
Amount of antibodies produced compared to that of the natural infection	8 times higher	100 times higher

Document 3

Part of the exercise	Exercise 2 Cervical Cancer and the HPV Virus	Grade 5 pts
1	Studies involving thousands of women suffering from cervical cancer shows that 75% of them have encountered the human papillomavirus (HPV) at some point of their sexual life. Thus this cancer is induced by a virus. The development of this cancer is slow it requires the persistence of the HPV infection for more than 13 years before inducing a genetic mutation at the origin of the cancer (document 1). Thus, this cancer is induced by a virus and its development needs time.	3/4
2	The environment: tobacco, nutrition... Status of the host: immune system, type of HLA	1/2
3	The specific immune response is cell mediated. Since the virus integrates it's DNA into the genome of infected cells and modifies their immunological self. This modified self is only recognized by the LT8 which are the effectors of the cell mediated response.	3/4
4	Document 2 shows that the highest percentage of cancers is 54,5% due to HPV 16 and 16% due to HPV 18. These percentages are higher than those of cancers induced by all the other types of HPV (more than 100 type of HPV). Thus the two types HPV 16 and HPV 18 are of high risk.	3/4
5	Both vaccines, Cervarix and Gardasil , requires the same amount and the same number of repetitions (3times) and immunize the body against the two types of HPV of high risk(HPV16 and 18). On the contrary, the level of produced antibodies induced by the vaccine Cervarix (100 times higher) is more important than that induced by the vaccine Gardarix that is 8 times higher than that produced in the case of natural infection. Thus the more efficient vaccine is Gardarix.	1
6	The antibodies produced due to vaccination neutralize the viruses before they bind to the membrane receptors of the target cells of the cervix and inhibit the viruses from infecting them. The viruses are thus eliminated (the formed immune complexes will be phagocytized by macrophages) and the lesions leading to cancer do not appear . Thus antibodies inhibit the HPV infection and protect the epithelium from genetic mutations that are at the origin of cervical cancer.	3/4
7	Get vaccinated before the first sexual intercourse. Do not smoke, have balanced healthy nutrition.	1/2

Exercise 2 (5 points)**AIDS and Treatments**

The human immunodeficiency virus (HIV) is responsible for the weakness of immune defenses in the organism, which leads to the death of affected persons. Document 1 shows the evolution of the concentration of T4 cells, measured in patients contaminated by HIV.

- 1- Analyze the results of document 1.
- 2- Draw out, from document 1, the cause of the appearance of opportunistic diseases.

In order to find a treatment that limits the consequences of opportunistic diseases, a series of studies is performed, some of which are represented below:

Study 1: Lymphocytes are removed from a monkey and B, T4 and T8 cells are separated.

- B cells are placed in chambers of culture 1 (1a, 1b and 1c) where molecules of antigen X are present at their bottoms. Only 0.01% of B cells remains fixed to the bottom of each chamber and is not eliminated by rinsing.
- T8 cells are placed in chambers of culture 2 (2a and 2b) where monkey cancerous fibroblasts are present at their bottoms. Only 0.01% of T8 cells remains fixed to the bottom of each chamber and is not eliminated by rinsing.
- Then, lymphocytes activated by the same antigens (X or cancerous fibroblasts) are added to certain chambers.

Document 2 shows the experimental conditions as well as the results.

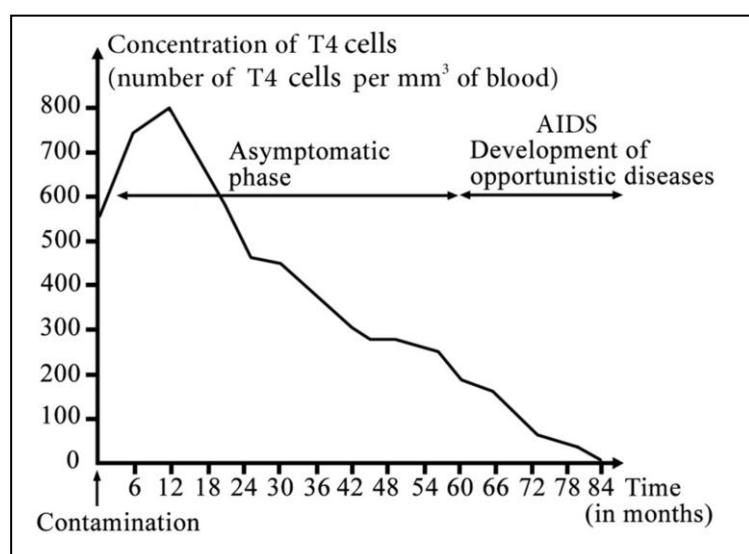
- 3- Interpret the results obtained in document 2.
- 4- Explain, by referring to all what precedes, the appearance of opportunistic diseases observed in document 1.

Study 2: Two groups of researchers have produced two treatments.

The first treatment is based on the principle of vaccination against some opportunistic diseases such as pneumonia. This treatment was tested on two categories of patients having a different number of T4 cells. The results are represented in document 3.

In the second treatment, three medicines are administered during 5 years to individuals whose number of T4 cells, at the beginning of treatment, is between 200 and 350 T4 cells/mm³ of blood. The results are shown in document 4.

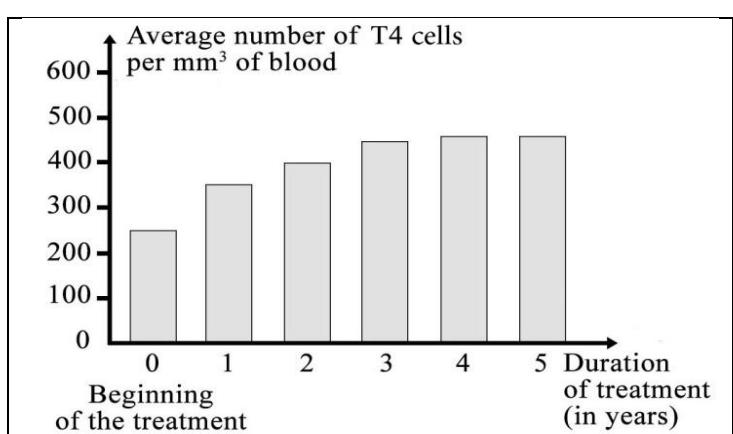
- 5- Explain the importance of vaccination.
- 6- Determine if the first treatment is efficient against the development of opportunistic diseases.
- 7- Show that the second treatment may delay the AIDS phase.

**Document 1**

Chamber	Existing lymphocytes	Added lymphocytes	Result
1a	B cells retained by antigen X	-	No antibodies
1b		Activated T4 cells	Presence of antibodies
1c		Activated T8 cells	No antibodies
2a	T8 cells retained by monkey cancerous fibroblasts	-	No lysis of fibroblasts
2b		Activated T4 cells	Lysis of fibroblasts

Document 2

Category	Average number of T4 cells/ mm ³ of blood	production of antibodies
1	> 500	Strong
2	< 200	Weak

Document 3**Document 4**

Part of the ex.	Exercice 2 AIDS and Treatments	Grade 5 pts
1	During the asymptomatic phase, the concentration of T4 cells/mm ³ of blood increases from 550 up to 800 just within 12 months after the contamination. On the contrary, this amount decreases from 800 to 200 at the 60 th month, the beginning of the appearance of opportunistic diseases until it becomes nil at the 84 th month.	1/2
2	The cause of appearance of opportunistic diseases is the low amount of T4 cells, less than 200/mm ³ of blood.	1/2
3	<p>The presence of antibodies is observed in culture medium 1b containing B lymphocytes activated by antigen X and LT4 activated by the same antigen. On the contrary, neither antibody are produced in culture medium 1a containing only B cells activated by antigen X or culture medium 1c containing B cells activated by the antigen and activated T8 cells. This implies that the cooperation only between T4 cells and B cells is indispensable for the secretion of antibodies.</p> <p>Lysis of monkey cancerous fibroblasts is observed in medium 2b containing T8 cells and T4 cells activated by the same antigen. On the contrary, no lysis is observed in culture medium containing only activated T8 cells. This implies that cooperation between T4 and T8 cells is indispensable for cellular lysis.</p>	1
4	<p>Document 2 shows the importance of T4 cells in the activation of specific humoral immune responses whose effectors are B lymphocytes and in cellular immune responses whose effectors are T8 cells.</p> <p>Document 1 shows that the opportunistic diseases appear when the concentration of T4 cells decreases to an amount inferior to 200/mm³. Thus, this low amount of interleukin secreted is insufficient to activate proliferation of activated B and T8 cells. This blocks specific immune responses and reduces general immunity of the organism, which renders the environment favorable to the development of opportunistic diseases.</p>	3/4
5	Vaccine ensures the first contact with this antigen and triggers immunological memory. Consequently, the body, after a second contact, develops a secondary response which is more amplified, more rapid and more durable against this antigen.	3/4
6	<p>In the first treatment, the vaccine isn't effective unless the amount of T4 cells/mm³ of blood is superior to 500 T4 cells/mm³ (document 3).</p> <p>But opportunistic diseases develop only when the amount is less than 200 T4 cells/mm³ (2.5 times less than 500). The first treatment is only efficient against pneumonia, one of the multiple opportunistic diseases. Thus this treatment isn't efficient against the development of the opportunistic diseases.</p>	3/4
7	The second treatment ensures the increase in the concentration of T4 cells in blood from 250 to 480 T4 cells/mm ³ between the beginning of treatment and the 5 th year (document 4). This doesn't lead to a concentration less than 200 T4 cells/mm ³ characterizing the AIDS phase which prolongs the asymptomatic phase and delays the AIDS phase.	3/4

Exercise 2 (5 points)**Conditions of LT8 Action**

The Choriomeningitic leukemia virus (CML) is slightly pathogenic and infects nervous cells. In the framework of studying the immune response against the infection by this virus, two experiments were performed.

Experiment 1: different viruses are injected into mice of different strains, Y and Z. The experimental conditions as well as the obtained results are shown in document 1.

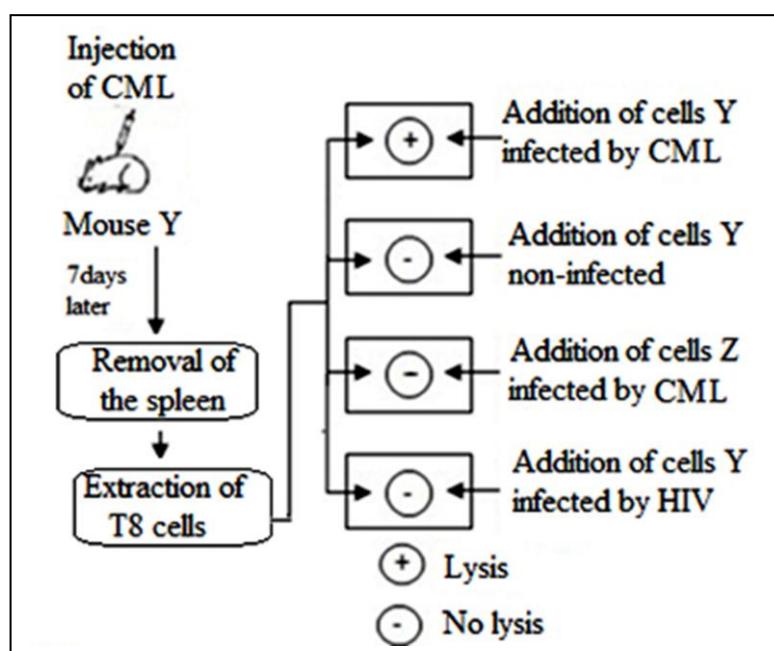
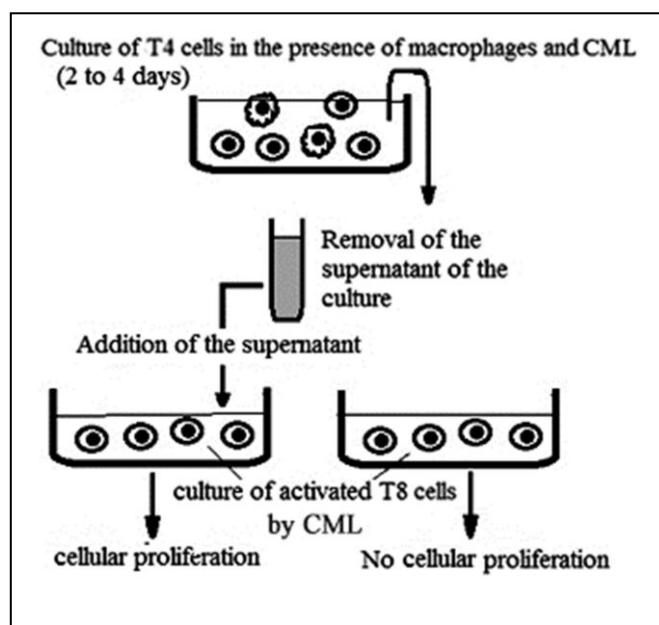
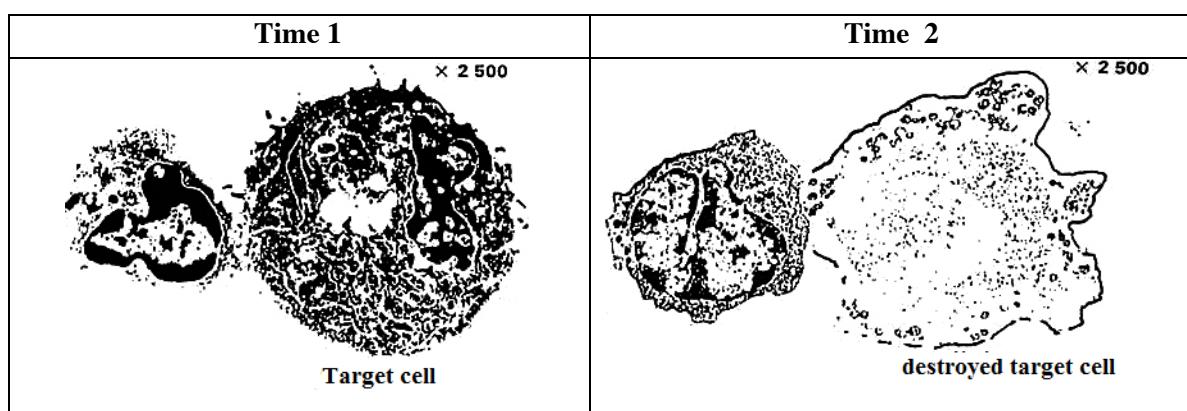
- 1- Name the specific immune response triggered against a virus and that triggered against a bacterium.
- 2- Interpret the results of experiment 1.

Experiment 2: T4 cells are cultured in the presence of macrophages and CML. The experimental conditions as well as the results are shown in document 2.

- 3- Determine the role and the mode of action of T4 cells as revealed in experiment 2.
- 4- Explain the role of macrophages in the culture of T4 cells in experiment 2.

Document 3 shows two electronographs, made at two successive times, of a target cell infected by CML in the presence of an activated T8 cell taken from experiment 2.

- 5- Draw a scheme showing the molecules involved in the recognition taking place between the activated T8 cell and the target cell.
- 6- Explain the mechanism shown in document 3.

**Document 1****Document 2****Document 3**

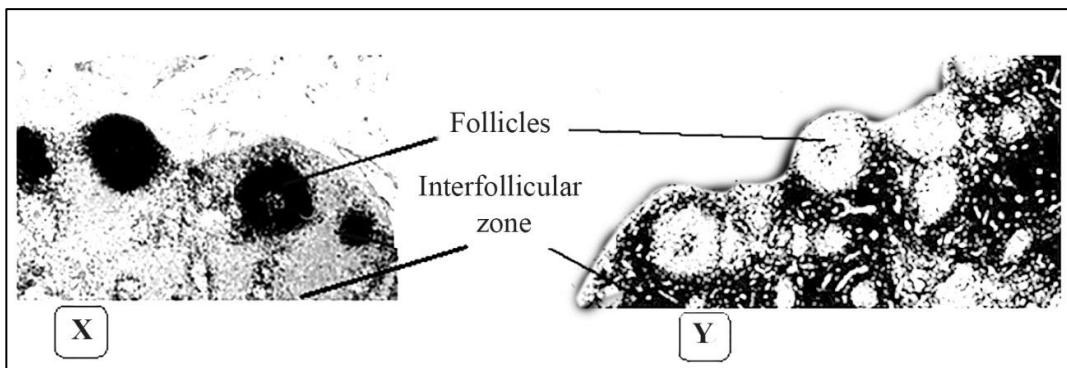
Part of the Ex	Exercise 2 (5 points)	Grade
1	<p>The response triggered against a virus is a specific cell mediated immune response.</p> <p>The response triggered against a bacterium is a specific humoral mediated immune response.</p>	1/2
2	<p>Cells Y infected by CML undergo lysis by T8 cells which are taken from mice of the same strain Y injected by CML. Whereas, cells of the same strain Y that are not infected don't undergo lysis. This shows that T8 cells destroy only infected cells.</p> <p>On the other hand, T8 cells which are taken from mice of strain Y injected by CML lyse the cells of same stain that are infected by CML, but they don't lyse cells of a different strain Z infected by the same virus CML. This implies that T8 cells lyse only infected cells that belong to the same strain.</p> <p>Cells Y infected by CML undergo lysis by T8 cells which are taken from mice of the same strain Y injected by CML. On the contrary, cells of same strain Y which are infected by another virus, HIV, are not lysed. This implies that T8 cells destroy only the cells infected by the same virus that activated them</p>	11/2
3	<p>There's only proliferation of T8 cells when we add the supernatant taken from a culture of T4 cells which are activated by CML in the presence of macrophages. Thus activated T4 cells stimulate the multiplication of T8 cells that recognized the same antigen, by secreting a substance, chemical messengers.</p>	3/4
4	<p>The macrophage phagocytizes and digests the CML virus, the obtained peptides get associated to HLA class II molecules and expressed at the cell surface. The macrophage becomes an antigen presenting cell APC. The APC fixes to T4 cells having specific receptors to the HLA- CML peptide complex thus activating the T4 cells leading to the formation of TH cells that secrete IL-2.</p>	3/4
5	<p>Scheme of the recognition site between T8 cells and the target cell</p> <p>The diagram illustrates the interaction between a T8 cell and a target cell. The T8 cell, at the top, has a T-cell receptor (TCR) labeled in blue. This TCR is shown binding to a specific protein complex on the surface of the target cell, labeled as the "HLA- CML peptide complex". The target cell is represented by an orange shape with a black oval inside. The interaction is depicted as a close proximity between the T8 cell and the target cell.</p>	3/4
6	<p>The T8 cell performs the double recognition by fixing to the HLA- CML peptide complex of the target cell (time 1). It secretes perforin molecules that form a channel through the plasma membrane of the target cell; then it releases granzymes that penetrate the target cell through the polyperforin channel leading to the degradation of its DNA and consequently to its lysis (time 2).</p>	3/4

Exercise 2 (4 points)**Hypertrophy of Lymph Nodes**

A temporary hypertrophy (swelling) of the lymph nodes is observed in an individual infected by an antigen like the tetanus toxin. In order to better understand the mechanisms involved in this hypertrophy, the following experiments are performed.

Experiment 1: The constituents of the lymph nodes of this individual are studied by using radioactive markers. Microradiographs are then performed.

The radioactive labeled zones appear in black on the microradiographs.

**Document 1**

Document 1 shows the results of labeled radioactive B lymphocytes (X) and of labeled radioactive T lymphocytes (Y).

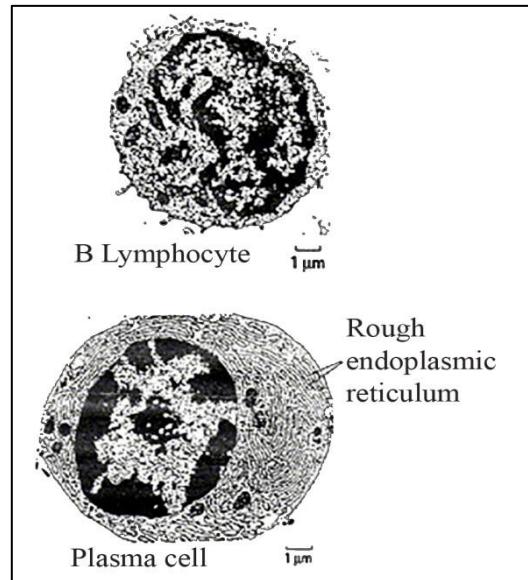
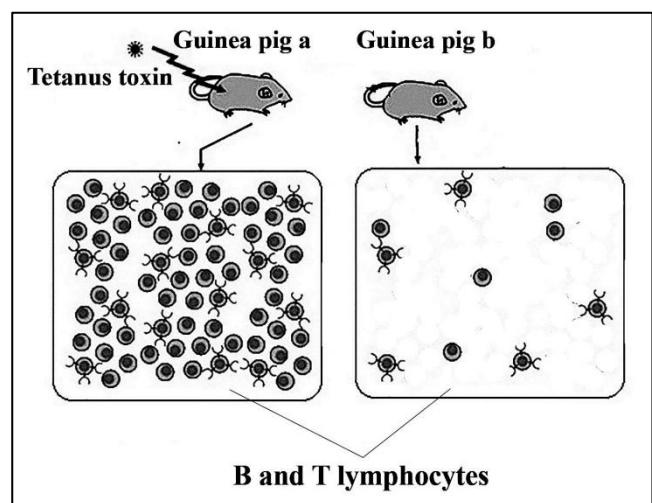
- 1- Deduce the localization of each lymphocyte population at the level of lymph nodes.

Document 2 shows microphotographs of the cells identified in the lymph nodes of the individual who is infected with tetanus toxin.

- 2- Specify the type of the immune response triggered against this antigen and revealed in document 2.
- 3- 3-1- Name the molecules secreted by this plasma cell.
3-2- Explain how the plasma cell is a cell adapted to the secretion of these molecules.

Experiment 2: cells are extracted from the lymph nodes of a guinea-pig (a) which is injected with tetanus toxin and from the lymph nodes of a healthy guinea pig (b). They are then purified to obtain only B and T lymphocytes. The results are schematized in document 3.

- 4- Interpret the results presented in doc 3.
- 5- Justify, referring to what precedes, the temporary hypertrophy of the lymph nodes observed in this individual.
- 6- Explain the role of TL involved in the immune response revealed in document 2.

**Document 2****Document 3**

Exercise 2 (5 points)**Roles of Macrophage**

The monocytes circulate in the blood and can migrate to the tissues where they become macrophages.

- 1-** Indicate the origin of monocytes.

In order to study the mode of action of macrophages and their cooperation with certain cells of the immune system, the following experiments are performed.

Experiment 1 :

Cells are extracted from the ganglia of a guinea pig which is immunized against antigen X. T4 lymphocytes and macrophages are isolated and placed in different culture media. The experimental conditions and the results are shown in document 1.

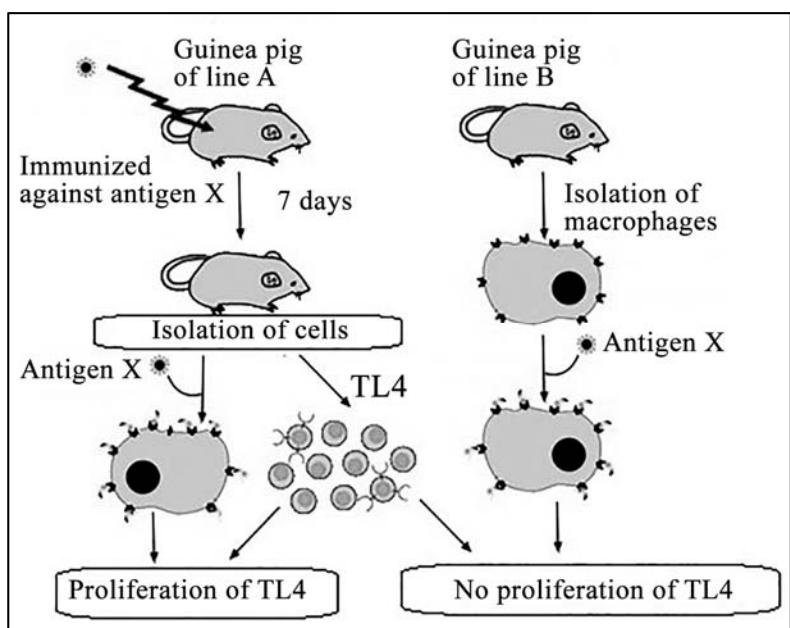
- 2-** Determine the conditions indispensable for the proliferation of T4 lymphocytes.

Culture medium	Conditions of the culture	Results
1	T4 lymphocytes and antigen X	No proliferation of T4 lymphocytes
2	T4 lymphocytes and macrophages	No proliferation of T4 lymphocytes
3	T4 lymphocytes, macrophages and antigen X	Proliferation of T4 lymphocytes
4	T4 lymphocytes	No proliferation of T4 lymphocytes

Document 1**Experiment 2 :**

An experiment is performed on two different strains of guinea pigs, A and B. The experimental conditions as well as the results are shown in document 2.

- 3-** Indicate the condition indispensable for the proliferation of T4 lymphocytes shown in this experiment. Justify the answer.

**Document 2****Experiment 3:**

Macrophages are incubated with the same antigen X labeled with radioactive ^{131}I odine isotope. At phase I, radioactivity is detected inside the macrophage, and in phase II a rapid degradation of antigen X is noticed. After some time 80% of radioactivity is detected in the culture medium in the form of ^{131}I odine isotope linked to peptides, while the remaining 20% are found attached to the cell surface.

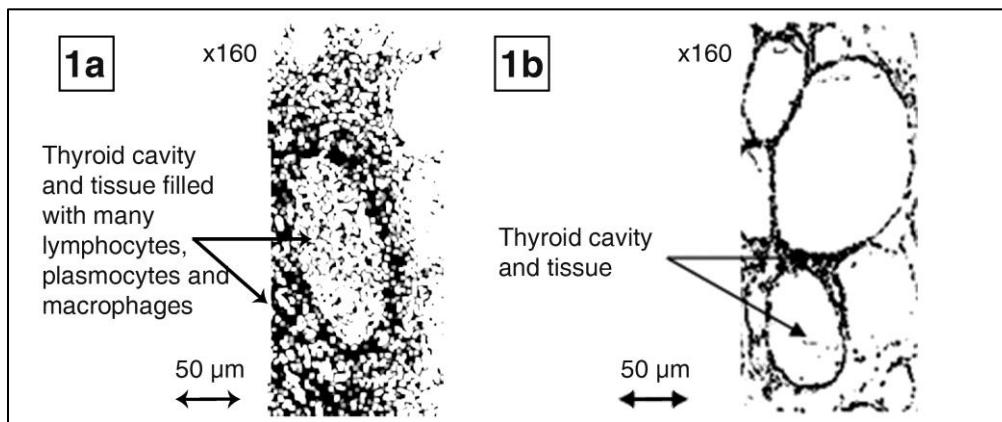
- 4-** Draw out the role of macrophages shown at phase I of experiment 3.
5- Explain the results obtained at phase II in experiment 3.
6- Explain the mode of action of the macrophages that permits the proliferation of T4 lymphocytes.
7- Specify the consequence of the absence of the macrophages on the specific immune responses.

Q	Exercise 2(5 Points)	Role of Macrophages	Mark
1-	Bone marrow		1/4
2-	Proliferation of T4 lymphocytes takes place only in culture of medium 3 in the presence of T4, macrophages and antigen X. Hence the proliferation of T4 lymphocytes necessitate the association or cooperation between T4 and macrophages in the presence of antigen X.		1
3-	The macrophages and the T4 cells must descend from the same strain. Since there is no proliferation of T4 lymphocytes when T4 cells of strain A are incubated with macrophages from another strain B. However proliferation takes place when T4 cells and macrophages previously in contact with an antigen X that descend from the same strain A.		1
4-	Phagocytosis		1/2
5-	At phase II of the experiment, 20% of radioactivity is detected on the surface of the cell, because a part (80%) of the degraded radioactive protein is eliminated out of the cell; The remaining 20% is degraded into peptides that are associated with MHC II on the surface of macrophages.		1/2
6-	The macrophages that are transformed into APC present the non-self-peptide associated with MHC II on its surface. So TCR of T4 lymphocytes bind to this complex and the T4 becomes activated.		3/4
7-	The induction of specific immune response ceases because the activation of T4 lymphocytes necessitate its binding to APC. So in the absence of activated T4 lymphocytes, no more secretion of interleukin 2 takes place which is responsible for launching the specific cell mediated immune response. Moreover, no interleukin 4 secretion takes place which is responsible for launching of the specific humoral immune response.		1

Exercise 2 (5 points)**A Case of Thyroiditis**

Sarah has a swelling of the neck at the level of thyroid gland and suffers from many troubles of metabolic origin. Blood analysis of Sarah shows that the concentration level of the thyroid hormones is noticeably lower than the normal values. The synthesis of these thyroid hormones necessitates the presence of a protein named thyroglobulin.

A biopsy is performed on the thyroid gland of Sarah. Document 1 represents the results of the microscopic observations of the sections of thyroid gland of Sarah (1a) and those of the normal thyroid gland (1b).

**Document 1**

- Formulate a hypothesis that can explain the results of biopsy of the thyroid gland of Sarah.

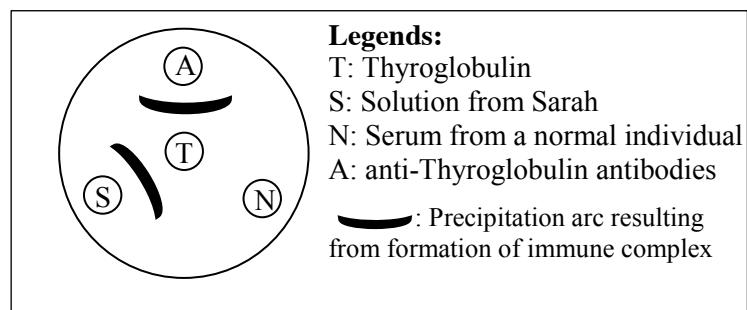
The immune and thyroid cells extracted from the thyroid gland of Sarah, are cultured in 3 different media. The conditions as well as the results are shown in document 2.

- Interpret the results shown in document 2.
- Identify the nature of the specific immune response revealed in document 2.
- Explain the following statement: "Macrophages induce specific immune response".

Culture	Cultivated Cells	Results
1	Thyroid cells + B Lymphocytes	Absence of antibodies
2	Thyroid cells + B Lymphocytes + Macrophages	Absence of antibodies
3	Thyroid cells + B Lymphocytes + Macrophages + T ₄ Lymphocytes	Presence of a large amount of antibodies

Document 2

Afterwards, immunodiffusion gel test is applied. A solution containing the protein thyroglobulin (T) is deposited in the central well, and three other different solutions are separately deposited in three peripheral wells: A solution of antibodies from Sarah (S), anti-thyroglobulin antibodies (A), and serum from a normal individual (N). The results are shown in document 3.

**Document 3**

- Show that Sarah suffers from an auto-immune disease directed against the self.

Exercice 2 (5 points)**Case of Thyroditis**

Q 2	Correction	Marks
1	Hypothesis : Sarah may have an infection in the thyroid gland. Sarah may have an auto-immune disease. Sarah may have a cancer at the level of the thyroid gland	
2	A large amount of antibodies is secreted in culture 3 in the presence of four types of cells : thyroid gland, B lymphocytes , macrophages and T4 lymphocytes. On the contrary, no antibodies are secreted in the absence of T4 lymphocytes (culture 2) and absence of macrophages (culture 1). This means that secretion of antibodies by B lymphocytes nécessitaires the presence of T4 lymphocytes and macrophages in the presence of an antigen, in this case the infected thyroid cells of sara.	0.5
3	Document 2 reveals the secretion of antibodies, therefore, the nature of specific immune response is humoral.	
4	When a macrophage phagocytoses and digests a cell or protein, resulting peptides are attached to HLA class II molecules and presented on the cell surface. The macrophage migrates to the closest lymph node, where it becomes an antigen presenting cell or APC. The T helper cells that are specific for the peptides presented by this APC remain attached to it. Then they are activated and they proliferate	
5	The anti-thyroglobulin antibodies in well A moves along the gel where it recognizes the thyroglobulin protein, fix to it and form an immune complex which appears as a precipitation arc. A similar precipitation arc (immune complex) is formed between well S and T which means that serum of sara contains antibodies specific to the protein thyroglobulin where they move along the gel and forms an immune complex . No such arc is revealed between well A and well N which lacks the anti-thyroglobulin antibodies since well N contains serum of a normal individual. Thus. Sara cells secrete antithyroglobulin antibodies which attack the thyroglobulin protein in her thyroid gland leading to problems in metabolism and swollen neck. This shows that Sara has auto immune disease.	

Exercise 2 (5 points)**Therapy against an autoimmune disease**

Type 1 diabetes (T1D) is due to an autoimmune disease. The current treatment that is based on insulin injection attenuates the symptoms of type I diabetes disease without curing it. For this reason, a research is carried out to verify the effectiveness of a new therapeutic approach to stop the progression of the autoimmune disease which is at the origin of this type of diabetes.

Measurements of the mass of certain components of the pancreas are performed during autopsies in healthy individuals and in individuals suffering from type 1 diabetes. Document 1 shows the obtained results.

1.1. Compare the obtained results.

1.2. Draw out the cause of type 1 diabetes.

The NOD mice (Non Obese Diabetic) develop a disease similar to T1D starting from the age of 10 weeks.

Document 2 represents islets of Langerhans of NOD mice at two different stages of diabetes: an early stage of diabetes (2 a) and a more advanced stage (2b). In this document, T8 lymphocytes appear in the form of black spots.

Note that these mice are not subjected to any viral infection.

2. Identify the type of the immune response involved in this autoimmune disease.

3. Explain the mode of action of T8 Lymphocytes on their target cells.

A new treatment for T1D is tested on two lots of NOD mice at the age of 4 weeks, before the onset of the disease:

- Lot A receives an injection of a saline solution that has no effect (control lot).
- Lot B is subjected to this new treatment.

Document 3 shows the occurrence of diabetes in these two lots of NOD mice.

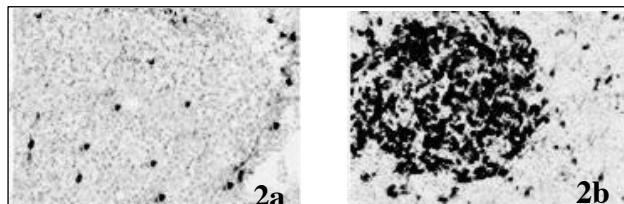
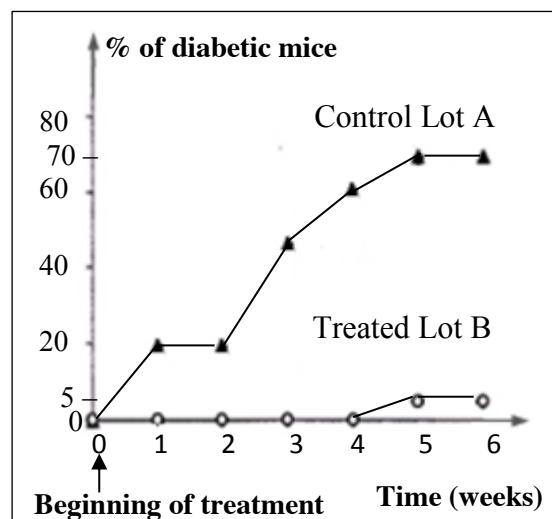
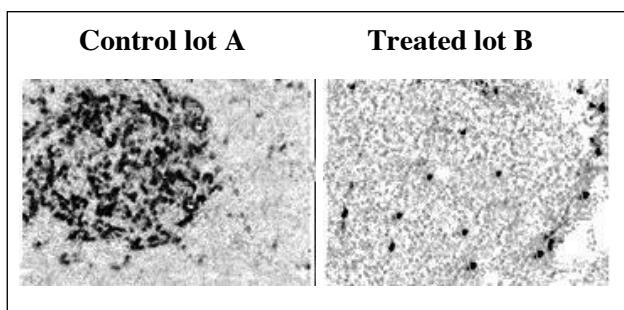
4. Draw a table representing the results of document 3.

5. Verify if this new treatment is effective against type 1 diabetes.

Document 4 presents the results of labeling cytotoxic T8 lymphocyte in the pancreas of beginning of treatment. T8 lymphocytes appear in form of black spots inside the islet of Langerhans.

6. Draw out how this new treatment slows down the progression of T1D.

	Healthy individual	Individual suffering from type 1 diabetes
Mass of the islets of Langerhans (mg)	1400	415
Mass of alpha cells (mg)	220	200
Mass of beta cells (mg)	850	0

Document 1**Document 2****Document 3****Document 4**

Q	Exercise 2 Therapy against an Autoimmune Disease	Mark																										
1.1	The mass of the islets of Langerhans in a healthy individual is 1400mg which is greater (3.38 times more) than 415mg in individual suffering from T1D. While the mass of alpha cells in a healthy individual is 220mg which is slightly greater than that of alpha cells in the affected individual (200 mg). However, the mass of beta cells in a healthy individual is 850mg which is greater than 0mg in the affected individual.	0,5																										
1.2	Type 1 diabetes is due to a lack of beta cells.	0,25																										
2	Document (2a) shows few T8 lymphocytes which appear in the form of black spots in islets of Langerhans of NOD mice at an early stage of diabetes. At a more advanced stage of diabetes (document 2b), the concentration of T8 lymphocytes represented by black spots in the islets of Langerhans of NOD mice increases. As T8 cells have a cytotoxic action against cells, these results show that beta cells are being attacked by T8 cells causing their disappearance in the individual affected by T1D (document 1). Since T8 cells are the effector cells involved in cell mediated immune response, then the immune response involved is specific cell mediated.	1																										
3	<p>During a cell mediated specific immune response:</p> <ul style="list-style-type: none"> - TL8 recognizes the antigenic peptides presented by MHC found on the membrane of target cells, through its TCR. - They are then activated by double recognition. - Once activated, and under the action of IL-2, T8 cells proliferate and form a clone. - Activated T8 cells differentiate into killer cells or cytotoxic TL which : <ul style="list-style-type: none"> • Secrete perforin which forms hollow channels through the plasma membrane of target cells. • Secrete granzymes that penetrate the polyperforin channels, leading to the degradation of its DNA. <p>This leads to apoptosis of target cells.</p>	1																										
4	<table border="1"> <thead> <tr> <th colspan="2">Time (weeks)</th> <th>0</th> <th>1</th> <th>2</th> <th>3</th> <th>4</th> <th>5</th> <th>6</th> </tr> </thead> <tbody> <tr> <td rowspan="2">% of diabetic mice</td> <td>Control Lot A</td> <td>0</td> <td>20</td> <td>20</td> <td>50</td> <td>60</td> <td>70</td> <td>70</td> </tr> <tr> <td>Treated Lot B</td> <td>0</td> <td>0</td> <td>0</td> <td>0</td> <td>0</td> <td>5</td> <td>5</td> </tr> </tbody> </table> <p style="text-align: center;">↑ Beginning of treatment</p> <p>Title: Table showing the variation of percentage of T1D in NOD mice with or without treatment, as a function of time.</p>	Time (weeks)		0	1	2	3	4	5	6	% of diabetic mice	Control Lot A	0	20	20	50	60	70	70	Treated Lot B	0	0	0	0	0	5	5	1,25
Time (weeks)		0	1	2	3	4	5	6																				
% of diabetic mice	Control Lot A	0	20	20	50	60	70	70																				
	Treated Lot B	0	0	0	0	0	5	5																				
5	<p>The results of document 3 show that the percentage of diabetic mice in control lot A (injected with a saline solution that has no effect) increases between 0 and 6 weeks from 0% to reach 70% which is a value much greater than that of treated lot B (subjected to the new treatment), where the percentage of diabetic mice remains null till 4 weeks, and then increases slightly to 5 % between 4 and 6 weeks.</p> <p>The new treatment has thus reduced the risk of developing type 1 diabetes, which confirms its effectiveness against this disease.</p>	0,75																										
6	This treatment seems to protect the beta cells of islets of Langerhans from the cytotoxic action of T8 lymphocytes; consequently slowing down the occurrence of type 1 diabetes in individuals at risk.	0,25																										

In the framework of studying the immune responses against the flu virus, several observations and experiments are performed.

First observation: Individuals who are infected by the flu virus show signs of an inflammatory reaction.

- 1- List the four signs of the inflammatory reaction.

Second observation: Document 1 presents the variation of the concentration of anti-flu antibodies as a function of time, following the infection by the flu virus.

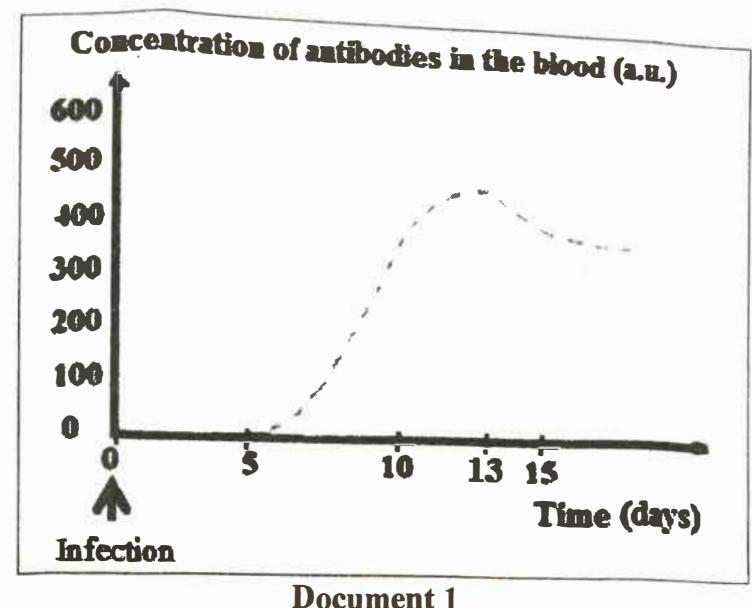
- 2- Identify the type of the specific immune response revealed by the results of document 1.

Experiment 1: The flu virus and anti-flu antibodies of increasing concentrations, C_1 , C_2 and C_3 , are added to different culture media containing human cells. The concentration of the infected cells is measured and the obtained results are presented in document 2.

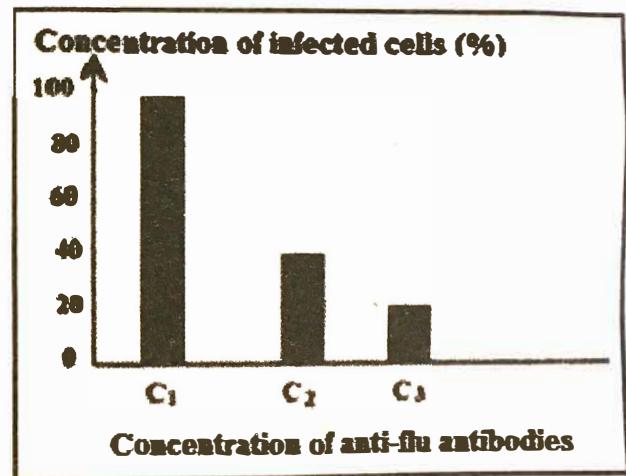
- 3- Interpret the obtained results.

Experiment 2: The action of antibodies does not permit the elimination of the cells infected by the flu virus. The monitoring of the number of cytotoxic T lymphocytes (Tc) and the infected cells in an individual infected by the flu virus shows the results presented in document 3.

- 4- Draw the graph showing the variation of the number of infected cells and that of Tc cells as a function of time.
- 5- Specify the type of the specific immune response revealed by the results presented in document 3.



Document 1



Document 2

Time (days)	0	3	7	9	13	15
Number of Tc cells	0	0	300	500	100	50
Number of infected cells	50	100	200	150	10	0

Document 3

Third observation:

Clinical observations show that the flu virus may be fatal in some individuals showing deficiency in T_H lymphocytes (case of AIDS).

- 6- Explain this observation.

Exercise 2 (5 points)**Graft and Immunological memory**

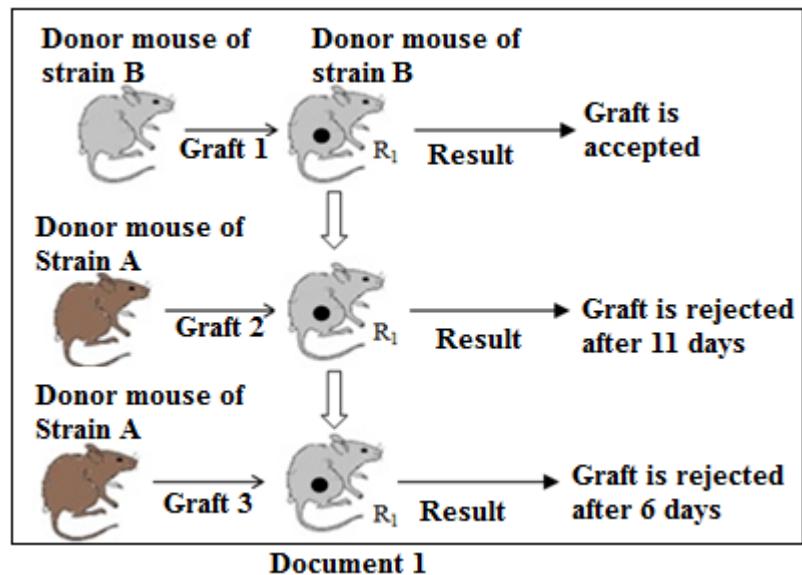
A study is performed to specify the mechanism of immunity involved in the rejection of skin graft in mice. Skin grafts are performed between different strains of mice, strain A and strain B. Document 1 shows the experimental conditions as well as the obtained results. The receiver mouse R₁ is the same in the three cases of grafting.

1. Interpret the obtained results.

In order to explain the results of the third graft, two hypotheses are proposed:

Hypothesis 1: Mice B possess memory T lymphocytes against the antigens carried by the cells of mice A.

Hypothesis 2: Mice B possess antibodies against the antigens carried by the cells of mice A.



Mice of strain B are hyper-immunized by grafting them for three times by, three weeks apart, by skin from mice of strain A. Then, the researchers extracted from these hyper-immunized mice of strain B serum (blood plasma) on one hand and lymphoid cells from lymphatic ganglia close to the graft on the other hand.

An experiment is performed on mice of strain B called “Nude” (named NB), which are not subjected to any prior treatment. The conditions and the results are shown in document 2.

Day 1 : Injection of mice NB	Day 3 : Grafts done on mice NB	Result
Serum from the hyper-immunized mice of strain B	Skin from mice of strain A	On Day 6: Acceptance of the graft On day 11: Rejection of the graft
Alive lymphoid cells from the hyper-immunized mice of strain B	Skin from mice of strain A	On day 6: Rejection of the graft
Dead lymphoid cells from the hyper-immunized mice of strain B	Skin from mice of strain A	On day 6: Acceptance of the graft On day 11: Rejection of the graft

Document 2**2. Verify, by referring to doc.1 and doc.2, which of the preceding formulated hypotheses is valid.**

The analysis of the lymphoid cells, responsible for graft rejection, present in the hyper-immunized mice gives the results presented in document 3.

3. Identify the cells X and Y in document 3.**4. Explain, by referring to all what precedes, the result of graft 3 in document 1.**

Hyper-immunized mice		
Lymphoid cells X	Lymphoid cells Y	
Percentage	95 %	5 %
Life Span	few days to few dozens of days	few months to few dozen of years
Proliferation	No	Yes

Document 3

Parts of ex	Exercise 2 (5 points)	Mark
1	<p>The skin graft is accepted when it is performed from a mouse of strain B to a mouse of the same strain B (graft 1). However, it is rejected after 11 days if the skin tissue is done between two mice of different strains: a donor mouse of strain A and a receiver mouse of strain B (graft 2).</p> <p>This shows that the graft is only accepted between individuals of the same strain.</p> <p>The graft rejection (graft 3) between two different strains A and B happens after 6 days, less than 11 days for graft 2 when the mouse of strain B has previously rejected the first skin graft issued from mouse of strain A.</p> <p>This shows that the immune response responsible for graft rejection is much faster during the second contact with the same antigen.</p>	1.5
2	<p>When serum from hyper-immunized mice of strain B is injected into "nude" mice (BN) of strain B followed by transplanting in them skin graft from mouse of strain A; 11 days later, the graft is rejected at the same duration as a control mouse in graft 2 in document 1, which has never been in contact with the antigen of mouse A.</p> <p>This means that, the serum of hyper-immunized strain B has no effect in the rejection of the graft.</p> <p>Therefore, the hypothesis, which states that mice B have antibodies which are at the origin of graft rejection, is invalid.</p> <p>When alive lymphoid cells taken from hyper-immunized mice B are injected into "nude" mouse of strain B (BN) then, followed by transplanting in them skin tissue from mouse A ; after shorter duration of time, 6 days later, the graft is rejected, similar to duration of time required by the mouse which receives the same graft for the second time, graft 3 of document 1. Moreover, the graft is always rejected at day 11 in the control mice of strain B that are injected by killed lymphoid cells taken from hyper-immunized mice of strain B.</p> <p>This means that the lymphoid cells are responsible for triggering response against the antigen.</p> <p>Hence, the hypothesis which states that mice B possess immune memory cells which are at the origin of graft rejection is valid.</p>	1.5
3	<p>Cells X are short-lived cells which life span range from days to tens of days and are involved in the cell-mediated immune response. Since differentiated immune cells have a short life span, hence these cells are the effector cells, Tc.</p> <p>Cells Y have a long life span, few months to tens of years, and they can proliferate. Since the cells having these characteristics are memory cells which appear after the first contact with the antigen, so cells Y are memory cells. And since this is a cell-mediated specific immune response, then cells Y are Tc memory cells.</p>	1
4	<p>Mice R1 of strain B develops a primary specific cell mediated immune response against graft A (graft 1). The activated Tc cells proliferate and give a clone of lymphocytes. Some of the daughter cells differentiate into "effector" cytotoxic Tc and others become memory cells specific against antigen A.</p> <p>After the second contact with the same graft A, memory Tc cells proliferate rapidly and differentiate to cytotoxic Tc, ensuring the rapid rejection of the graft.</p> <p>Since, the triggered secondary immune response is faster, the rejection of the skin tissues in graft 3 of document 1 is obtained after 6 days instead of 11 days.</p>	1

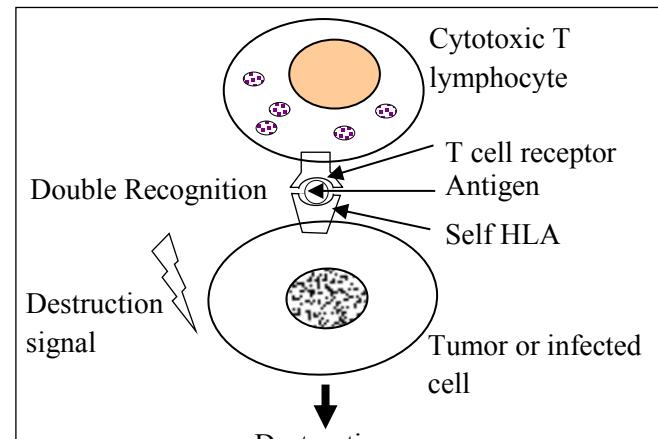
Exercise 2 (5 points)**Pregnancy and Immune Defense**

The fetus is a kind of temporary allograft that survives for nine months. However, fetal cells should be non-self for the mother's immune system because they express protein markers different from those of the mother. A research is performed to discover some mechanisms that allow the fetus to escape the mother's immune system during pregnancy.

The fetus is surrounded by a tissue called trophoblast, which isolates it from the maternal immune system.

The trophoblast cells do not express HLA class I proteins that are mainly involved in the cytotoxicity of certain lymphocytes against the non-self (Document 1).

1. Explain the mechanism of cellular cytotoxicity of Tc lymphocytes.
2. Determine the cause of ineffectiveness of Tc lymphocytes against the fetal cells.

**Document 1**

Moreover, the trophoblast cells carry on their surface and secrete in the medium a protein called HLA-G, a non-polymorphic molecule. A hypothesis assumes that this HLA-G protein prevents trophoblast cells from being recognized by the immune system as non-self-cells.

In order to validate this hypothesis, experiment 1 is performed. The conditions and the results of this experiment are presented in document 2.

Experiment 1:

Medium	A	B	C
Conditions	Immune cells of the mother	Immune cells of the mother	Immune cells of the mother
	Non-self cells	Trophoblast cells carrying HLA-G molecules	Trophoblast cells carrying HLA-G molecules blocked by a chemical substance
Results	Lysis of the non-self cells	No Lysis of trophoblast cells	Lysis of trophoblast cells

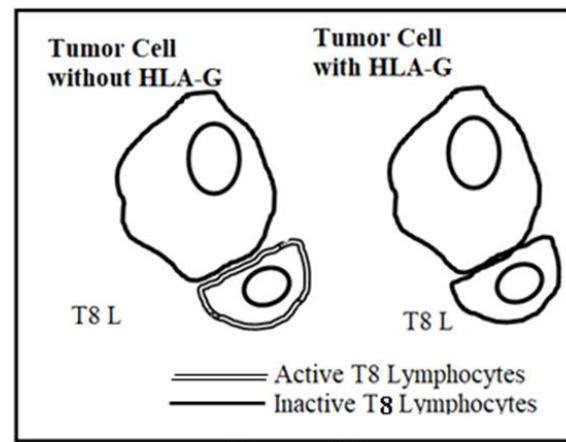
Document 2

3. Do the obtained results validate the tested hypothesis? Justify the answer.

Some cancer cells also produce HLA-G protein. In an attempt to find out if this molecule allows the cells to escape the action of the T lymphocytes, the following experiments, 2 and 3, are performed.

Experiment 2: Macrophages are placed in contact with the non-self cells carrying HLA-G. Their capacity to activate T4 lymphocytes becomes reduced.

Experiment 3: T8 lymphocytes are cultured in the presence of two types of cancer cells. The results are shown in document 3.

**Document 3**

4. Determine, by referring to each of the experiments 2 and 3, how the HLA-G contributes to making the specific immune response less effective.

Ex	Part	Exercise 2 (5 points) Pregnancy and Immune Defense	Mark
2	1	The T cell receptor (TCR) on the membrane of Tc lymphocyte binds to HLA-I – nonself-peptide complex on the target cell membrane, tumor or infected cell. Then, Tc lymphocyte releases its perforin molecules which assemble into polymers that form a hollow channel through the target cell membrane. Then, the Tc releases granzymes that penetrate into the target cell through the polyperforin channels triggering an enzymatic chain reaction within the cell leading to DNA degradation. This causes cell death by apoptosis.	1.5
	2	The action of Tc lymphocyte on the target cell necessitates the double recognition of the nonself peptide associated to self HLA-I protein. The trophoblast isolates the fetus from the maternal immune system. The cells of this trophoblast do not express the self HLA- class I proteins; thus, these cells are not recognized by Tc lymphocyte. This makes Tc lymphocyte unable to reach and destroy the fetal cells.	1
	3	The hypothesis is validated since the mother's immune cells lyse the non-self cells (medium A) but not the trophoblast cells that carry HLA-G molecules (medium B). This shows that HLA-G prevents the action of maternal immune cells on trophoblast cells. This is also confirmed by the result obtained in medium C, where the trophoblast cells carrying blocked HLA-G molecules are lysed by maternal immune cells.	1
	4	Experiment 2 shows that the capacity of macrophages to activate T4 lymphocytes is reduced when placed in contact with the non-self cells carrying HLA-G . Furthermore, the activation of the T4 cells is a step necessary for the induction of the specific immune response, humoral and cellular mediated. Hence, this immune response becomes less effective. The results of experiment 3 show that T8 lymphocyte remains inactive when it binds to a tumor cell which carries HLA-G molecules. However, it becomes active if HLA-G molecules are absent. Thus, T8 lymphocyte are not activated by tumor cells carrying HLA-G molecules. Hence, the specific immune response triggered by lymphocytes is less effective.	1.5