**Results:**

|  |  |  |
| --- | --- | --- |
| Species | neostigmine IP 0.005%, 0.1ml/10g | atropine 0.4% 0.1ml/10g OP/ IP |
| Mouse | After the intraperitoneal injection  About 7 minutes: The mice developed muscle weakness, convulsions, and drooling | After the mice were given gavage  At about 43 minutes, the mice showed more activity than the saline group, but the symptoms were not very significant.  After about 51 minutes: the mice were very hyper, became difficult to hold, held the cage upside down, and could observe changes in the abdominal cavity, breathing significantly more and more frequently. Then we put them to death, and we can see the exophthalmos of the mice |
| Rabbit | The rabbits were injected with neostigmine at a dose of 2mL/kg for about 1 minute and 50 seconds. The rabbit dies. |  |

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| adsorption | Saline 1 | Neostigmine 1 | Saline 2 | Neostigmine 2 |
| A：background | 0 | 0 | 0 | 0 |
| B：control group | 0.372 | 0 | 0.52 | 0.368 |
| C：experimental group | 0.143 | 0.172 | 0.11 | 0.243 |
| ACHE enzyme activity（U/mL） | 61.56 | 47.72 | 78.845 | 33.965 |

**Discussion:**

**Neostigmine** primarily acts by **inhibiting** **acetylcholinesterase (AChE)** to increase the concentration of **acetylcholine (ACh)**, rather than directly promoting an increase in AChE concentration. This was clearly seen in the controlled experiment. CHE activity in the serum of the saline group was significantly higher than that of the Neostigmine group. This mechanism leads to the accumulation of acetylcholine at the neuromuscular junction and other affected areas, thereby enhancing acetylcholine signal transmission. As an important neurotransmitter, acetylcholine functions through interactions with two major types of receptors: nicotinic receptors (N receptors) and muscarinic receptors (M receptors). Neostigmine mainly produces effects in the peripheral nervous system, especially at the neuromuscular junction. By increasing the concentration of acetylcholine, it enhances activation of N receptors, which leads to increased muscle contraction strength. It can also affect M receptors, particularly in tissues controlled by the parasympathetic nervous system, such as the gastrointestinal tract and heart, potentially causing side effects such as increased salivation and slowed heart rate.

So for the Neostigmine injected mice, we observed strong muscle contractions, most likely caused by activation of the N receptor. In the case of rabbit, he died after injection, and we speculated that ACh overactivated M receptor due to high dose, leading to heart failure and thus death.

**Atropine** primarily acts by **blocking muscarinic receptors** (M receptors), affecting both the peripheral and central nervous systems. In the peripheral nervous system, atropine reduces saliva and sweat secretion, increases heart rate, dilates pupils, and slows down gastrointestinal motility. In the central nervous system, it may cause side effects such as hallucinations or confusion.

So after injecting atropine for some time, we observed that the mice were abnormally excited, and it was more difficult to control than before. When we finally killed the mouse, its heart still had a faint beating. This is likely because atropine blocks the M receptor, which prevents ACh from binding effectively, causing the mice's hearts to beat faster and cause other overreactions.

**Conclusion:**

Our findings highlight two main points. Firstly, neostigmine effectively inhibits acetylcholinesterase (AChE), decreasing AChE activity and thereby increasing acetylcholine (ACh) concentration. This increase in ACh concentration allows for greater binding to muscarinic receptors, resulting in various physiological responses. As a result, neostigmine has been shown to be effective in slowing excessive heart rate and preventing muscle cramps.

Secondly, regarding atropine, while we are unable to elucidate its mechanism of action at the molecular biological level, it is hypothesized to interact with muscarinic receptors, likely by inhibiting them. This inhibition may contribute to an increased heart rate, making atropine a potential treatment for heart failure. However, further research is needed to fully understand atropine's effects and mechanisms.