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Research article

Expression and role of cystatin C in hyperthermia-induced brain injury in rats

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Abstract: Cystatin C, the full name of cystatin C, is one of the most potent cathepsin inhibitors currently known, which can strongly inhibit cathepsin in lysosomes and regulate the level of intracellular proteolysis. Cystatin C plays a very broad role in the body. High temperature-induced brain injury leads to very serious damage to brain tissue, such as cell inactivation, brain tissue edema, etc. At this time, cystatin C can play a crucial role. Based on the research on the expression and role of cystatin C in high temperature-induced brain injury in rats, this paper draws the following conclusions: high temperature can cause very serious damage to the brain tissue of rats, which can seriously lead to death. Cystatin C has a protective effect on brain cells and cerebral nerves. When the brain is damaged by high temperature, cystatin C can relieve the damage of high temperature to the brain and protect brain tissue. In this paper, a detection method for cystatin C with more outstanding performance is proposed, and compared with the traditional detection method, the detection method in this paper is verified to have more accurate accuracy and excellent stability through comparative experiments. Compared with traditional detection methods, it is more worthwhile to use and is a better detection method.

Keywords: cystatin C; rat brain injury; high temperature damage; brain injury

1. Introduction

Cystatin C, an inhibitor of cysteine proteases, acts on the lysosomal system to regulate the

activity of a variety of protein hydrolases. Under oxidative stress, cystatin C significantly promotes cell survival: it strongly inhibits histone protease activity, protects the stability of intracellular membrane structures, promotes autophagy and prolongs cell life by modulating molecular signaling pathways. The structure of cystatin C can inhibit the enzyme of certain proteins, he shows the re-stacking composition of the protein, and can maintain the original structure. Dimerization can undergo structural changes, resulting in a single protein structure. Cystatin C is gradually dimerizing, and it will endanger human health, especially the elderly. Aggregates can be obtained in other ways, and their unfolded molecules are infinite [1]. Cystatin C is an essential glycosylated protein produced at a constant rate by all nucleated cells tested. It is freely filtered through the glomerulus and catabolized primarily in the renal tubules (it is not excreted or absorbed intact). Serum cystatin C is an indicator of increased glomerular filtration rate (GFR) relative to serum creatinine because it is independent of age, sex, and muscle mass. Cystatin C is different from other GFR. By comparison, we found that the concentration of cystatin C is more flexible and sensitive than other GFR markers [2]. Serum cystatin C concentration was inversely correlated with glomerular filtration rate, as or better than serum creatinine, indicating that it is continuously formed and cleared as the main source of extracellular fluid. But based on the data we know now; it is not known how well the GFR matches it [3]. We have demonstrated that serum cystatin C measurement is a more selective change indicator. But we now know very little about cystatin C concentrations is still sparse. Therefore, our main subjects are adults. We draw blood from volunteers participating in the experiment, and then measure the concentration of cystatin and creatinine. When people are under the age of 50, nearly 90% of people have cystatin C in the range of 0.51–0.93 m [4].

At present, we know that if the kidney function of an older person has problems, then his lifespan will be affected. It happens that cystatin C can prevent related problems, solve problems related to kidney function, and prolong the life of the elderly. This has a very important development guide for the future of medicine [5]. After the brain is injured, other diseases will occur one after another, and serious damage to the nerves will occur. We know that the related anti-cancer genes can help the nerves in the brain recover faster and slow down the damage to the brain after brain injury, but we don't know how it is done yet. Western blot and immunohistochemical's data and analysis of PIDD can let us know that PIDD will slowly increase after brain damage, and then slowly decrease again. And the place with the most PIDD is in the neuron [6]. We found several chemical mediators in the rat brain, which are formed as a result of cerebral edema. We carried out experiments on rats, put the rats in a high temperature environment, and then extracted brain tissue from the rats after brain injury, and examined the brain edema of the rats and the changes in some cells. Indomethacin and other drugs can slow down brain damage in rats to varying degrees. Experiments have also shown that brain damage caused by high temperature is very complex and only related to nerves [7]. After rat brain injury, by extracting rat brain tissue, it was found that IL-1 can share the use of molecules with the rat brain tissue extract. After brain injury in rats, IL-1 becomes very active and reproduces very quickly, which can help the rapid production of antibodies in rats, which can restore brain injury in rats, an important role in cell repair after injury [8].

Recent evidence suggests that progesterone treatment attenuates rats and can cause many other diseases after brain injury, but we don't know whether it is related to age, so we experimented with old rats, let them receive the vehicle, and exercised to observe the recovery. And two days later, the degree of cerebral edema was checked, and it was found that the symptoms of the treated rats were alleviated. It was also shown to reduce apoptosis, reduce swelling and improve exercise performance

over time in the 16 mg/kg group [9]. At present, the main discussion of brain injury caused by ischemia is the toxicity of calcium. According to previous reports, although it is known that calcium can protect animals with from ischemia, experiments have not been able to prove this point of view. Protein blocks calcium from passing through. channel, but this can also keep the amount of calcium in a balanced state [10]. So far, the temperature of the earth has been gradually rising every year, and it will increase at a faster rate in the future, which is a cause for concern and a very serious problem. Plants experience fluctuations in ambient temperature, and many animals are warm-blooded animals, their life and metabolism are closely related to temperature, and they can control their own temperature. Therefore, as the global temperature increases, the impact on plants will be more obvious, and they will suffer very serious damage. It is estimated that crop yields decrease by 17% for every 1 °C increase in average growing season temperature [11]. Adaptation of livestock to high ambient temperatures often results in lower yields as animals reduce their metabolic rate and feed intake to accommodate the increased heat load. Ideally, one would like to select both increased yield and thermal resistance to increased thermal loads. This will require both identification and selection to improve heat dissipation and production mechanisms [12]. To judge whether an animal can withstand high temperatures, the environment and details of the experiment are very important. In the experiment, if the animal is directly placed in a high temperature environment, the experimental results are not very accurate, because it is ignored that under normal circumstances, the temperature is gradually increased. As a result, the experimental environment is different from the real ecological environment, and when the temperature gradually increases, the animals endure the high temperature for a significantly longer time. Flies exposed to faster rates are more tolerant to heat knockdown [13].

In our daily life, in our living environment, the temperature is an unavoidable problem, so the relevant experimental data and reports about high temperatures are very important, such as continuous high temperature exposure experiments. In order to complete related experiments, many rapid heating methods have been invented, such as heating with a fire source, heating with hot water, or heating by wrapping the human body. It is to know the relationship between the change in temperature and the time its temperature lasts. Normally, the higher the temperature and the longer the time, the stronger the animal's response. Therefore, it can be proved that the longer the high temperature exposure time, the more serious the damage caused by the high temperature will be due to high temperature and adrenocorticotropic hormone on the content of ascorbic acid glands of guinea pigs, rabbits and albino rats were studied [14]. When an animal is suddenly placed in a warmer place, there is less ascorbic acid in the animal's liver and other organs, but gradually returned to a normal level in the following 24 hours. Only a few percent are found in the liver, kidneys, and spleen. Though the high temperature, the rate of decline in adrenal is reduced to a lower degree, which may be due to the animal's acclimation or adaptation to the high temperature environment [15]. Cystatin C exhibits variability in different organisms, with somatic expression in response to temperature in animals in different environments. Cystatin C has very limited effect and unsatisfactory efficacy in the expression of cystatin C in brain injury in rats. Therefore, the search for effective prevention and treatment methods to reduce the occurrence of cerebral ischemic injury, alleviate post-ischemic neurological dysfunction and promote its clinical application is a challenge that needs to be addressed.

2. Materials and methods

2.1. The role of cystatin C

Cystatin C, a member of the cystatin superfamily, is a secreted protein. Genes are domestic genes. Cystatin C synthesis is not tissue-specific, all eukaryotic cells express and still secrete cystatin C. Cystatin C is present in the body fluids of the human body and will not be affected by other causes. Cystatin C is mainly metabolized in the kidney, where it is almost completely filtered by the glomerulus and absorbed and completely degraded in the proximal tubule. The kidney is the only organ that removes cystatin C from the circulation, so the level of cystatin C in serum primarily determines GFR. Cystatin C can be considered as an endogenous substance reflecting ideal GFR. The humoral cystatin C in normal adults is shown in Figure 1.

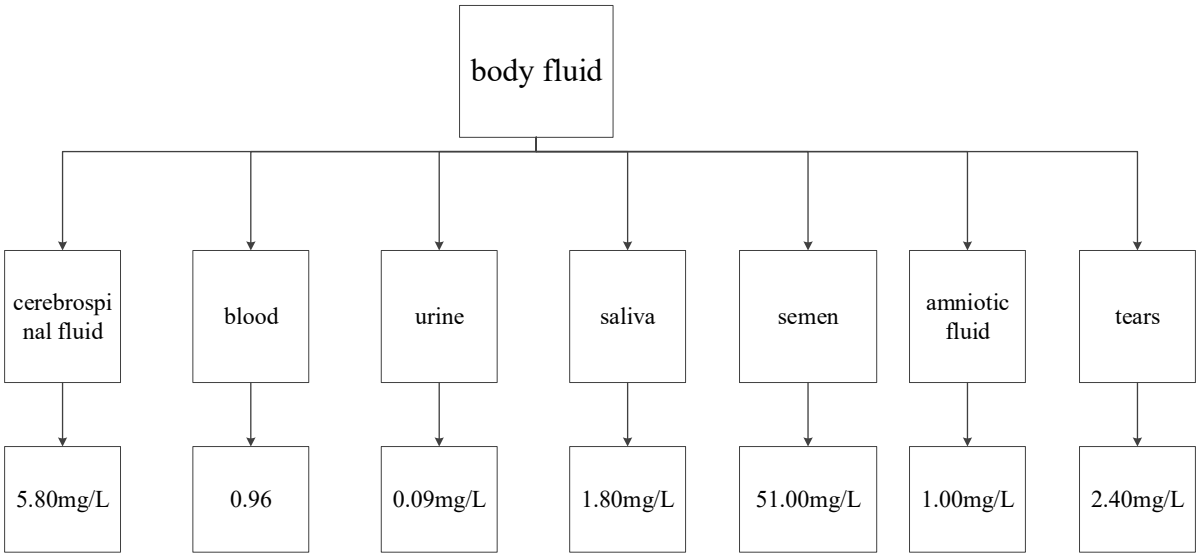

Figure 1. Distribution of cystatin C in human body fluids.

The relative molecular weight of cystatin C is 13,400, its component is nitrogenous acid, and there are 120, cystatin C is a basic secreted protein. The nuclei throughout the human body can synthesize and secrete cystatin C, which is mainly found in extracellular fluids, including cerebrospinal fluid, blood, sperm, and other human body fluids, including cerebrospinal fluid and cerebrospinal fluid. It is also expressed in other cells such as neurons. More than 99% of cystatin C in the human body is freely filtered through the glomerulus and reabsorbed into renal tubular epithelial cells for degradation, so that almost all cystatin C is metabolized in the kidneys.

Cystatin C exists in the cells and body fluids of humans and other mammals, and can help organisms carry out normal physiological activities. Cystatin C controls the rate of proteolysis inside and outside the cell. Studies have shown that cystatin C participates in cell proliferation and differentiation by regulating the activity of intracellular lysosomal cathepsins, regulating the synthesis and metabolism of intracellular proteins, and participating in biological activities by supporting the digestion and metabolism of viruses and bacteria by immune system cells. Antiviral and antibacterial activity; acts on TGF-β receptors in positive cells and tumor cells, inhibits TGF-β

signaling pathway, inhibits its binding activity, and participates in the proliferation and metastasis of tumor cells; regulates cathepsin K and participates in the decomposition of bone matrix and absorption.

1) Neuroprotection: The addition of exogenous cystatin C to cell culture media increases cell viability under stress and decreases cell viability up to a range of concentrations. Cytotoxic damage, such as energy consumption and oxidative stress, significantly increases cell survival. 2) Protection of cerebrovascular: According to existing research, cystatin C may cause diseases related to cerebrovascular. When the gene for cystatin C is mutated, the walls of blood vessels in the brain can become blocked, causing blood vessels to rupture and bleed, leading to fatal strokes in early life. This type of stroke is called hereditary vascular disease, amyloid cystatin C. The increased amyloid activity of cysteine C increases the production of amyloid fragments in brain tissue, which can cause atherosclerosis, while cystatin C slows and prevents atherosclerosis. Studies have shown that serum cystatin C levels in patients with atherosclerosis are inversely correlated with disease severity. Cystatin C plays a very important role in the formation and development of atherosclerosis. Some researchers have suggested that the change of serum cystatin C concentration may become a benchmark for the diagnosis of acute myocardial infarction and stroke to a certain extent. However, some clinical studies have shown that higher serum cystatin C levels are associated with higher mortality and cardiovascular disease in the elderly, suggesting that excessive cystatin C levels may have adverse effects on the cerebrovascular system.

When the body temperature exceeds 41 °C. The human body will have a high fever, accompanied by obvious symptoms such as seizures, coma, shock, and bleeding. Elevated body temperature will speed up metabolism, increase the decomposition of substances, generate more heat, and form a vicious circle. If body temperature exceeds 41 °C, cells in the body's solid organs, especially brain cells, may degenerate, leading to seizures, convulsions, coma, heart failure and respiratory failure. Some enzymes may become inactive, causing permanent damage to brain cells, and leading to death.

2.2. Cystatin C detection methods

Cystatin C detection methods are divided into two types: heterogeneous detection and homogeneous detection. Heterogeneous detection includes single immunodiffusion (SRID), enzyme immunoassay (ELISA), radioimmunoassay (RIA) and fluorescence immunoassay (FIA) four detection methods; homogeneous detection includes particle counting immunoassay, immunotransmission turbidimetry (PETIA) and immune nephelometry (PENIA) three detection methods. Heterogeneous detection is complicated and time-consuming, and it is inconvenient to be used in clinical practice. Homogeneous detection has a strong advantage in time, and is not easily interfered by other factors, and at the same time, the recovery rate of homogeneous detection is high. The comparison of detection methods of cystatin C is shown in Figure 2.

Typical cystatin C sensor acquisition systems use photodetectors for signal conversion, so the received signal is very weak. To ensure accurate results, many processing circuits, such as differential amplifiers, are designed in hardware. To ensure accuracy, in terms of software, the average filtering algorithm is designed in hardware. Mediation is used to remove the influence of uncontrolled factors such as the environment on the outcome. The wavelet thresholding method is also used to align the measurement results so that the measurement value is closer to the true value. The steps and structure of the acquisition system of the cystatin C detection instrument are shown in Figure 3.

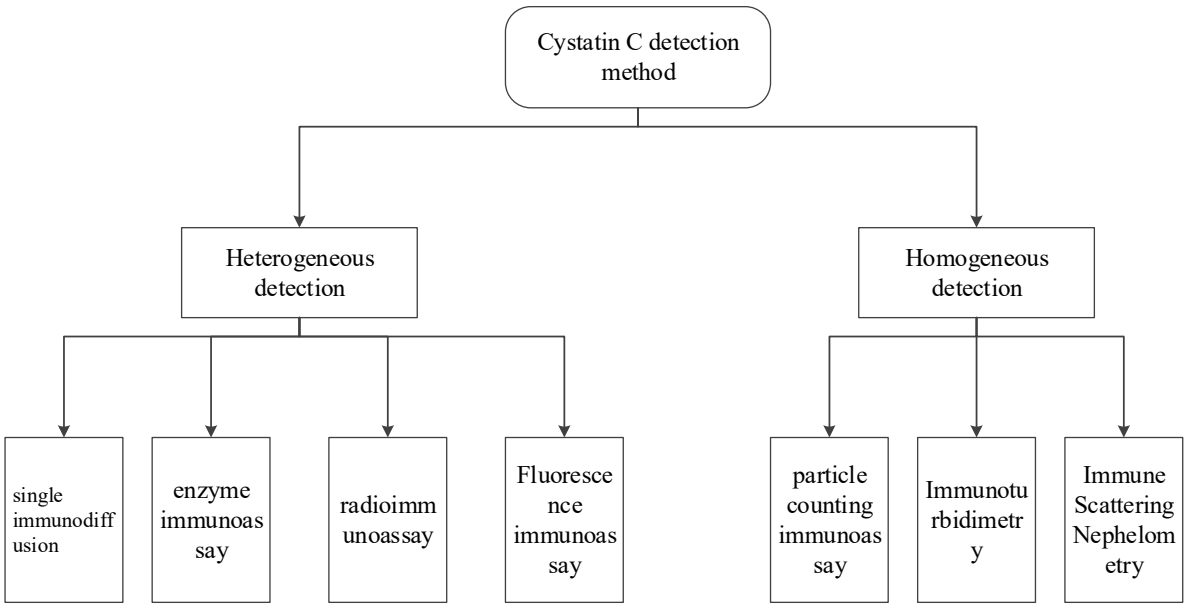

Figure 2. Cystatin C detection method.

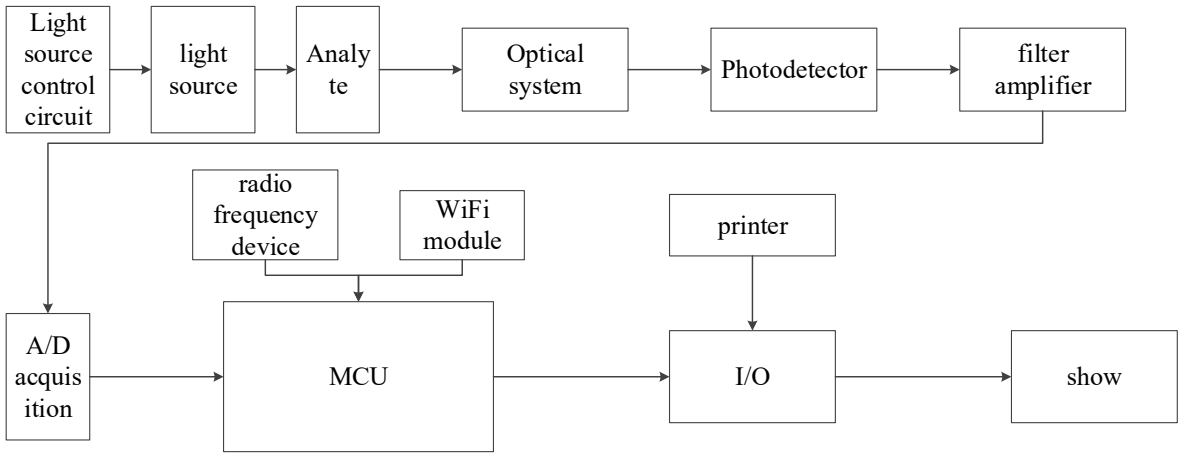

Figure 3. Operation diagram of cystatin C detector.

3. Results

3.1. Turbidimetry

When the light beam passes through the cuvette containing the suspension, the relationship between the intensity of the transmitted light and the intensity of the incident light will be attenuated to a certain extent due to the absorption and attenuation of the medium itself. As shown in the following formula.

$$I_T = I_0 e^{-(a+s)l} \tag{1}$$

$$I_T = \left(\frac{I_0}{I_T}\right) = A = ln(\alpha + s)\, l \tag{2}$$

If the definition of A in the formula (1) is the absorbance of the suspension, then.

$$A = log\left(\frac{V_0}{V_T}\right) \tag{3}$$

The accuracy and bias for the test can be calculated using the following equations.

$$r = \frac{\sum[(x_i - x)(y_i - y)]}{\sqrt{(x_i - x)(y_i - y)^2}} \tag{4}$$

$$B = \frac{M - T}{T} \times 100\% \tag{5}$$

3.2. Nephelometric method

When the reaction in the test tube progresses to a certain extent, it will no longer change. At this time, if the diameter of the particles in the test tube is larger than the wavelength of the light emitted by the light source, the light signal emitted by the light source will be weakened. The measured particles show a certain functional relationship, and the content of the sample to be tested is calculated through the functional relationship, so it is also called turbidimetry. The expression is as follows:

$$I = I_0 e^{-\tau b} \tag{6}$$

where I represents the transmitted light intensity, I_0 represents the incident light intensity, b represents the optical diameter, and τ represents the turbidity. The value of I depends on b, and the larger the value of b, the smaller the value of I.

A relationship like Beer's law can be obtained from the above formula:

$$log\,1\,0^{p0/p} = kbc \tag{7}$$

Since the particles in the reagent are very small, when the scattering phenomenon occurs, the magnitude of the light intensity satisfies Rayleigh's law:

$$I_S = \frac{9\pi^2}{\lambda^4 r^2}\left(\frac{n_2^2 - n_1^2}{n_2^2 - 2n_1^2}\right) v^2 N I_0 \left(\frac{1 + cos^2\,\theta}{2}\right) \tag{8}$$

Its simplified form is defined.

$$I_S = KNI_0 \tag{9}$$

3.3. Rayleigh scattering

When the particle size is smaller than the incident wavelength, the relationship between the scattered light intensity of a single particle and the scattering angle θ can be expressed by the following formula.

$$I(\theta) = I_0 \frac{9\pi^2 v^2}{\lambda^4 r^2}\left(\frac{n_2^2 - n_1^2}{n_2^2 + 2n_1^2}\right)^2 \left(\frac{1 + cos^2\,\theta}{2}\right) \tag{10}$$

It is assumed that the scattering of different particles does not cancel each other and that the

intensities of light scattered by the particles may overlap. At this time, the sum of the scattered light intensity N of the particles per unit volume can be taken as N times the scattered light intensity of a single particle, which can be expressed as:

$$I(\theta) = NI_0 \frac{9\pi^2 v^2}{\lambda^4 r^2} \left(\frac{n_2^2 - n_1^2}{n_2^2 + 2n_1^2}\right)^2 \left(\frac{1+cos^2\theta}{2}\right) \tag{11}$$

When viewed in the vertical direction of the incident light, it can be defined as:

$$I(90°) = kNI_0 \tag{12}$$

where k is recorded as:

$$k = \frac{9\pi^2 v^2}{\lambda^4 r^2} \left(\frac{n_2^2 - n_1^2}{n_2^2 + 2n_1^2}\right)^2 \tag{13}$$

where k is denoted as the scattering coefficient.

3.4. Mie scattering theory

As the particle size increases when the particle size is larger than the wavelength of the incident light, the Rayleigh scattering theory is no longer valid, the proportional relationship between the intensity of the scattered light and the frequency of the incident light no longer exists, it lasts longer, and the intensity of the scattered light is clarified:

$$I_M = K_M A N I_0 \tag{14}$$

In the formula, I_M is defined as the scattered light intensity, K_M is defined as the Mie scattering coefficient, and A is defined as the surface area of the scattering source particle. The larger the value of I_M, the stronger the intensity of scattered light.

The intensity of the light generated by scattering is proportional to the concentration of the particles, and can be written as:

$$I_S = K_S N I_X \tag{15}$$

In the formula, I_S is defined as the scattered light intensity, K_S is defined as the scattering coefficient, N is the solution concentration, and I_X is the incident light intensity. The value of I_S determines the value of the entire formula.

The above formula can also be expressed as

$$V_S = KNV_0 \tag{16}$$

3.5. Establishment of scattering model

According to the Mie scattering theory, the scattered light intensity at a point P in the scattered light field can be written as

$$I(r, \theta, \varphi) = \frac{\lambda^2 I_0}{4\pi^2 r^2} [i_1(\theta)\, sin^2\, \varphi + i_2(\theta)\, cos^2\, \varphi] \tag{17}$$

Similarly, the scattering amplitude function is expressed as:

$$S_1 = \sum_{n=1}^{\infty} \frac{2n+1}{n(n+1)} \{a_n \pi_n(cos\,\theta) + b_n \tau_n(cos\,\theta)\} \tag{18}$$

$$S_2 = \sum_{n=1}^{\infty} \frac{2n+1}{n(n+1)} \{a_n \tau_n(cos\,\theta) + b_n \pi_n(cos\,\theta)\} \tag{19}$$

where a_n, b_n is the Mie scattering coefficient, which is expressed as:

$$a_n = \frac{\psi_n(a)\,\psi_n'(ma) - m\psi_n'(a)\,\psi_n(ma)}{\xi_n(a)\psi_n'(ma) - m\xi_n'(a)\,\psi_n(ma)} \tag{20}$$

$$b_n = \frac{m\psi_n(a)\,\psi_n'(ma) - \psi_n'(a)\,\psi_n(ma)}{m\xi_n(a)\psi_n'(ma) - \xi_n'(a)\,\psi_n(ma)} \tag{21}$$

And

$$\pi_n(cos\,\theta) = \frac{P_n^{(1)}(cos\,\theta)}{sin\,\theta} \tag{22}$$

$$\tau_n(cos\,\theta) = \frac{d}{d\theta} p_n^{(1)}(cos\,\theta) \tag{23}$$

Here is the relative refractive index of the cystatin C antigen-antibody immune complex particle and the mixed solution, and the value is about m = 1.25.

$$\psi_n(z) = \sqrt{\frac{\pi z}{2}} J_{n+\frac{1}{2}}(z) \tag{24}$$

$$\xi_n(z) = \sqrt{\frac{\pi z}{2}} H^{(2)}_{n+\frac{1}{2}}(z) \tag{25}$$

4. Discussion

In order to further explore the specific situation of high temperature-induced brain injury and the expression and role of cystatin C in high temperature-induced brain injury in rats. We carried out a series of experiments to verify and analyze the effects of high temperatures on rats through the limb performance and brain tissue composition of rats at different temperatures. At the same time, the changes and effects of cystatin C were observed, to further verify and discover the expression and effect of cystatin C in high temperature-induced brain injury in rats.

4.1. Experimental materials and experimental content

In this experiment, 84 rats of the same age and weight will be selected from an animal center, and the 84 rats will be randomly divided into groups to verify the content of brain tissue components in different environments. High temperature damage to rat brain tissue. Then grouped, extracted rat brain tissue fluid, and analyzed the changes in brain tissue water content (BWC), Evans Blue (EB) and other components in the rat brain tissue fluid. At the same time, a cystatin C experiment was established. group to observe and study the role of cystatin C in high temperature-induced brain injury.

4.2. Experiments related to brain injury in rats caused by high temperature

In the experiment, 54 rats were randomly selected from 84 rats for grouping, 28 rats were randomly selected as the control group, and 28 rats were placed in an incubator for normal activities. Another 28 rats were placed in a high temperature box of 42 degrees, and two groups of rats were placed in the box for 48 hours, and 4 observation time points were set at 12 h, 24 h, 36 h and 48 h. And 14 rats in each group were planned as subgroups for the detection of Evans blue content in brain tissue. After 48 hours, the rats in the two groups were sacrificed, and brain tissue samples were extracted from the rats. For brain water content detection Evans blue content detection, etc. The specific experimental groups and the survival of rats are shown in Table 1.

Table 1. Experimental grouping and survival of rats.

Group	Number	Temperature	12 h Deaths	24 h Deaths	36 h Deaths	48 h Deaths
Test group	28	42 ℃	0	3	5	10
Subgroup 1	14	42 ℃	0	1	3	4
Control group	28	25 ℃	0	0	0	0
Subgroup 2	14	25 ℃	0	0	0	0

During the experiment, we found that the experimental animals in the experimental group became very impatient, and then gradually lost their vitality and even died. During this period, the mice in the experimental group developed eyeball hypertrophy and edema, and some mice developed corneal necrosis. The skull was cut open, and it was found that the brain tissue was edema, cerebral vascular occlusion, meningeal adhesion, enlarged gyri and superficial groove, and the left side was more prominent than the right side. There was no significant change in the control group. The changes in brain water content in rats are shown in Figure 4.

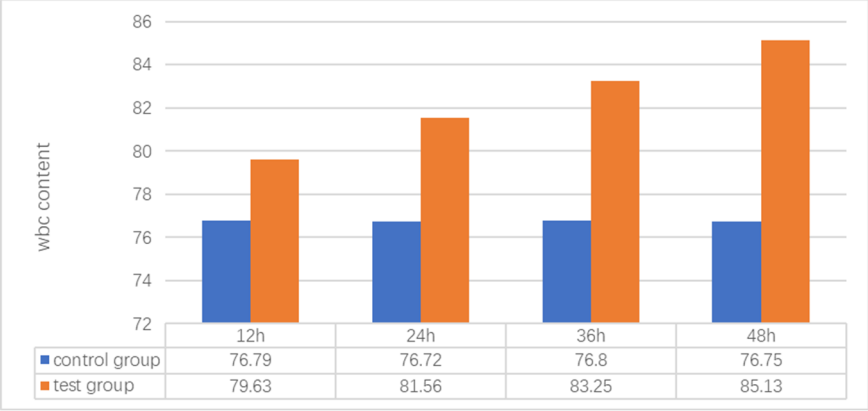

Figure 4. Changes in brain water content in rats.

From the data in Figure 4, it can be seen that the brain water content of the rats at 25 °C did not change much, but the brain water content of the rats at a high temperature of 42 °C gradually increased, which means that at 42 °C The metabolism of rats in high temperature is accelerated, which gradually causes hypoxia, which in turn causes cerebral edema, brain tissue swelling, cerebral

vascular congestion, cell swelling, vacuolar degeneration, and even necrosis. It shows that high temperature has serious damage to the brain tissue of rats, which can cause serious brain damage.

Compared with the rats in the control group, the Evans blue content in the brain tissue fluid of the rats exposed to a high temperature of 42 °C also increased significantly, and became higher and higher as time went on. The specific experimental results are shown in Figure 5.

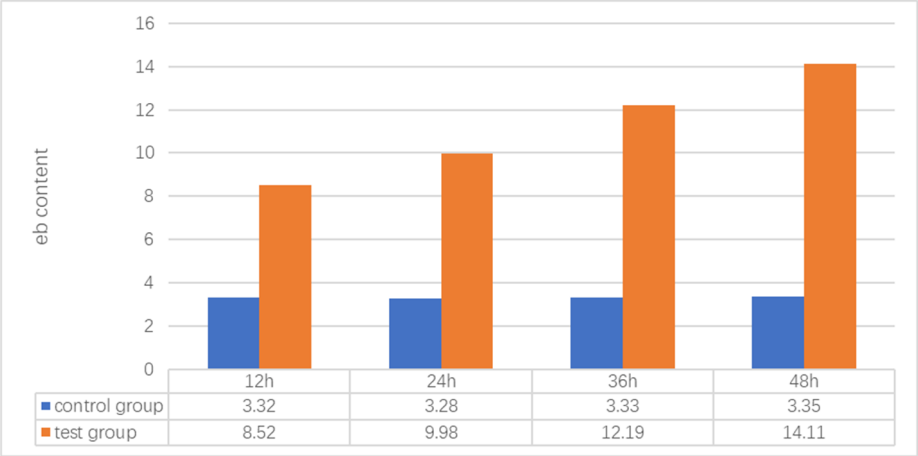

Figure 5. EB changes in rats.

4.3. Experiments related to the effect of cystatin C on brain injury induced by high temperature in rats

In this experiment, 84 rats in the experimental materials were randomly divided into three groups, each group was assigned 28 rats, and the three groups were: 1) The control group: 28 rats were placed in a constant temperature of 25 °C; 2) Control group 2: 28 rats were placed in a high temperature chamber at 42 °C without any treatment; 3) Experimental group: 28 rats were placed in a high temperature chamber at 42 °C, and injected an appropriate amount of cystatin C into the rats.

Two groups of rats were placed in the box for 48 hours, and 4 observation time points were set at 12 h, 24 h, 36 h and 48 h. And 14 rats in each group were planned as subgroups for the detection of Evans blue content in brain tissue. After 48 hours, the rats in the two groups were sacrificed, and brain tissue samples were extracted from the rats. For brain water content detection Evans blue content detection, etc.

After the rats were sacrificed, when the brain water content in the extracted brain tissue fluid was detected, it was found that the brain water content of the experimental group was higher than that of the control group 1, but it significantly decreased compared with the control group 2. This indicated that cystatin C played a significant protective role. The specific experimental data are shown in Figure 6 below.

From the data in Figure 6, it can be seen that the rats in the three groups have the lowest brain water content in the 25 °C incubators, and there is not much fluctuation in the brain water content. The highest is the control group 2, indicating that the brain tissue of the rats in the control group 2 has been severely damaged, resulting in brain damage. Although the brain water content of rats in the experimental group was higher than that in experimental group 1, it was significantly decreased compared with the control group 2. This indicates that cystatin C plays a role in protecting the brain tissue of rats from high temperature-induced brain injury.

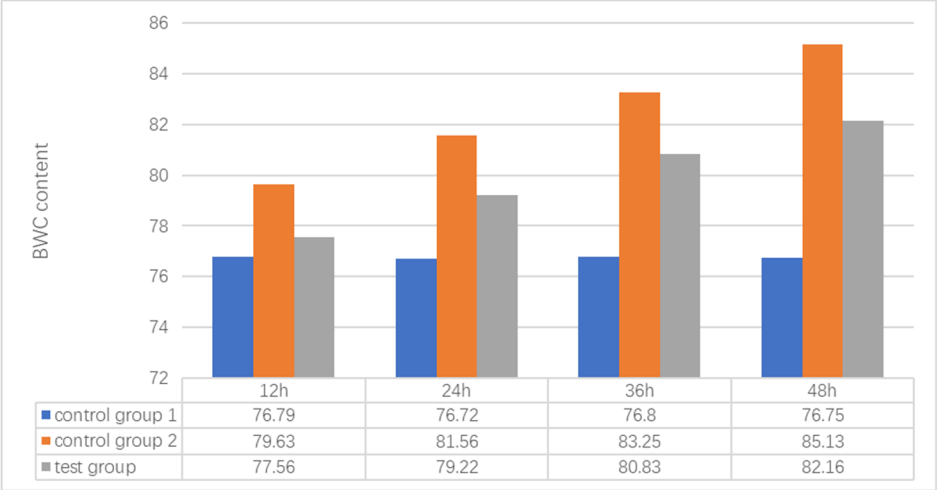

Figure 6. Changes in BWC content of rats in different groups.

Compared with the rats in the control group 1, the Evans blue content in the brain tissue fluid of the rats exposed to a high temperature of 42 °C also increased significantly, and it became higher and higher with the passage of time. Although the content of Evans blue also increased, it decreased significantly compared with the control group 2. The specific experimental data are shown in Table 2.

Table 2. Changes of rat EB content in different groups.

Group	12 h	24 h	36 h	48 h
control group 1	3.32	3.28	3.33	3.35
control group 2	8.52	9.98	12.19	14.11
test group	6.24	7.68	9.82	11.95

4.4. Comparative experiment of cystatin C detection methods

There are many detection methods for cystatin C. Heterogeneous detection is complicated and time-consuming, which is inconvenient to be used in clinical practice. Homogeneous detection has a strong advantage in time and is not easily interfered by other factors. The advantages of high phase detection recovery rate. We have compared the main common detection methods with the detection methods used in this paper in many aspects. The specific data are shown in Table 3.

Table 3. Comparison of cystatin C detection methods.

Method	CV	Measurement time	Reference range	shortcoming
SRID	11	38 h	1.3 ± 0.26	complex operation
ELISA	10–12	16 h	1.1 ± 0.42	long detection time
RIA	none	16–21 h	0.96 ± 0.20	polluted
FIA	none	3 h	none	expensive
Method	3–5	5 min	< 1.25	none

In order to further explore the accuracy of the detection method in this paper, we used the

algorithm in this paper to repeat the determination of 10 groups of cystatin C solutions with the same concentration at different times, and judged the accuracy of the detection method by the dispersion coefficient. The specific experimental data are shown in Table 4.

Table 4. Detection accuracy of detection methods for different concentrations of cystatin C.

Concentration	Detection times	Mean	Standard Deviation (mg/L)	CV (%)
0.237	10	0.256	0.01025	4.003
0.774	10	0.795	0.03188	4.010
1.363	10	1.387	0.06002	4.327
2.740	10	2.711	0.10586	3.905
5.500	10	5.516	0.21860	3.963
9.650	10	9.628	0.43422	4.510

Different cystatin C assays have different accuracy, and accuracy defines whether the assay has outstanding performance and whether it is worthwhile to use. This experiment will verify whether the algorithm in this paper has outstanding performance compared with other traditional methods through comparative experiments. With the increase of cystatin C concentration, the detection method can extract cystatin C more clearly, but the extraction accuracy cannot be guaranteed. In this experiment, three traditional methods and the method in this paper are used to extract and compare cystatin C. The specific experimental results are shown in Figure 7 below.

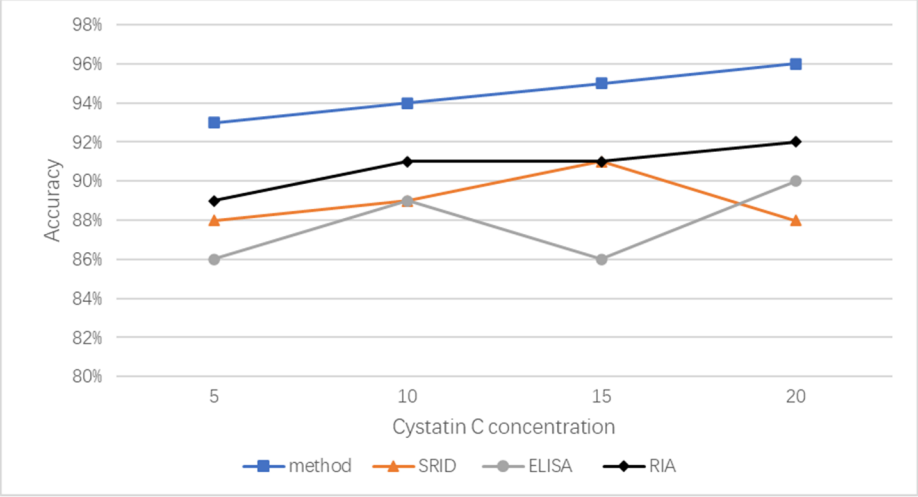

Figure 7. Accuracy comparison of detection methods.

From the data in Figure 7, it can be seen that the detection method in this paper has higher detection accuracy than other traditional detection methods, and the accuracy gradually increases as the concentration increases, while the accuracy of the traditional detection method is relatively low and does not steadily increase as the concentration increases. This shows that the detection method and algorithm in this paper have extremely high accuracy, which proves that the detection method in this paper has outstanding performance compared with the traditional detection method, and is a more worthwhile detection method for cystatin C.

Whether the detection method has excellent performance is generally judged by the absorbance

of the detection method to cystatin C during the detection. The higher the absorbance, the better the performance. In this experiment, the performance of the algorithm was judged by comparing the absorbance of different detection methods in the detection of different concentrations of cystatin C. The specific experimental data is shown in Figure 8 below.

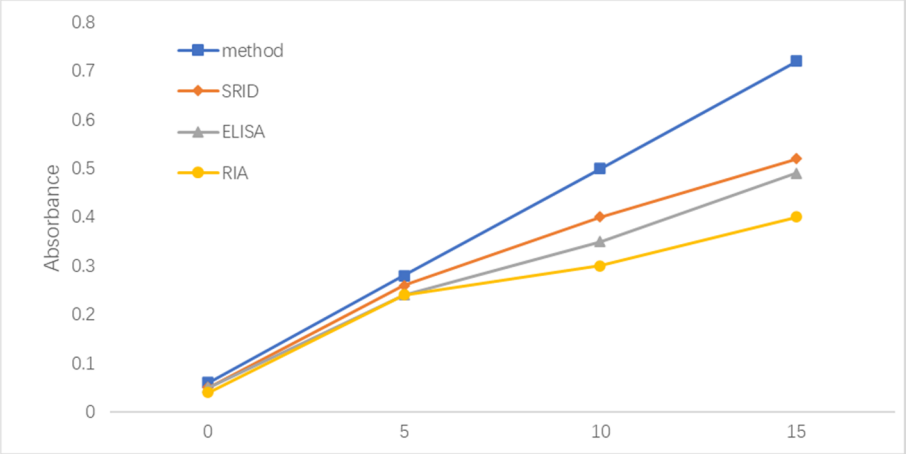

Figure 8. Performance comparison of detection methods.

Analysis of the data in Figure 8 in this paper shows that the absorbance of the cystatin C detection method in this paper is higher than that of other traditional detection methods, and it can complete the task better when detecting cystatin C. And the absorbance can be increased stably and efficiently with the increase of the concentration of cystatin C, while other traditional detection methods not only have low absorbance, but also cannot increase stably in the follow-up. This shows that the detection method in this paper has a very strong performance.

Whether the performance of the detection method is excellent, its stability is an important reference point. If the algorithm is very stable and efficient, then the performance of the algorithm is very outstanding. In this experiment, the stability of the algorithm is judged by the stability of the algorithm as the concentration changes when the detection method is detected. And using the standard deviation as a quantitative evaluation index, the experimental results are shown in Table 5.

Table 5. Stability comparison of detection methods.

Detection Method	Standard Deviation
Our Method	0.005
SRID	0.036
ELISA	0.024
RIA	0.018

It can be seen from the analysis of the data in Table 5 that the detection method in this paper has the smallest standard deviation, and the SRID method has the largest standard deviation. Since the smaller the standard deviation value, the higher the stability of the algorithm, it proves that the detection method in this paper has better stability and more outstanding performance than other traditional cystatin C detection methods, and is a more worthwhile algorithm to use.

5. Conclusions

Cystatin C is indispensable in living organisms, it can help evaluate kidney function, and can play a protective role in the brain. In the event of brain injury caused by high temperature, it can reduce the damage to brain tissue and protect the safety of brain tissue. Cystatin C also has a variety of detection methods. With the rapid development of the times, the detection methods of cystatin C are constantly improving. The cystatin C detection method proposed in this paper, combined with the characteristics of modern optoelectronics, enables the detection of cystatin C to be completed more efficiently and accurately. Make high-performance detection methods more worthy of use and promotion. At the same time, the role of cystatin C in brain protection and its clinical application has a very important impact.

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Conflict of interest

The authors declare there is no conflict of interest.

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