Triglyceride(TG) Content Assay Kit

Note: Take two or three different samples for prediction before test.

Operation Equipment: Spectrophotometer

Catalog Number: NA0819

Size:50T/48S

Components:

Reagent I: Self-provided reagent, add 45 mL of n-heptane and 45 mL of isopropyl alcohol to a glass bottle. Seal and mix well, storage at 4℃.

Reagent II: 7 mL×1 bottle. Storage at 4℃.

Reagent III: 30 mL×1 bottle. Storage at 4℃.

Reagent IV: 10 mL×1 bottle. Storage at 4℃, protected from light.

Reagent V: 30 mL×1 bottle. Storage at 4℃, protected from light.

Reagent VI: 30 mL×1 bottle. Storage at 4℃, protected from light.

Standard: powder ×1 bottle, add 5 mL of Reagent I before use. 1mg/mL triglyceride standard solution, storage at 4℃.

Product Description:

Triglyceride(TG) is a fat molecule formed by long-chain fatty acids and glycerol, which is not only the main component of cell membrane, but also an important respiratory substrate. The TG is extracted with isopropyl alcohol, then hydrolysis to glycerol and fatty acids after saponification of TG by KOH. Glycerol is oxidized by periodic acid to form formaldehyde. Condensation of formaldehyde and acetylacetone to form yellow components in presence of chloride ions. The yellow component has a characteristic absorption at 420 nm and proportional to the TG content.

Reagents and Equipment Required but Not Provided:

Spectrophotometer, water bath, adjustable pipette, 1 mL glass cuvette, n-heptane, isopropyl alcohol, distilled water, 100 mL empty glass bottle.

Procedure:

I. Sample preparation

1) Tissue: Ice-bath homogenate is conducted according to the ratio of tissue mass (g): Reagent I volume (mL) = 1: 5~10 (it is suggested to take about 0.1 g of tissue and add 1 mL of Reagent I). Centrifuge at 8000 g for 10 minutes at 4℃, supernatant is used for test.
2) Bacteria or cell: Suggested 5 million with 1 mL of Reagent I. Splitting bacteria or cell with ultrasonic for 1 minute (power 20%, work time 2 s， interval 1 s). Centrifuge at 8000 g for 10 minutes at 4℃, supernatant is used for test.
3) Serum: Detect directly.

II. Procedure:

1. Preheat Spectrophotometer for 30 minutes, adjust the wavelength to 420 nm, set zero with distilled water.
2. Preheat water bath to 65℃.

Reagent name (μL)	Blank tube (A_B)	Standard tube(A_S)	Test tube(A_T)
Distilled water	200	-	-
1 mg/mL Standard solution	-	200	-
TG test solution	-	-	200
Reagent I	625	625	625
Reagent II	125	125	125
Mix thoroughly after adding Reagent I, add Reagent II, shake strongly for 30 s, stand several minutes. After layering, 75 μL of the upper layer solution is taken and put it into a new EP tube.			

3. Detect TG content:

Reagent name (μL)	Blank tube (A_B)	Standard tube(A_S)	Test tube(A_T)
Upper layer solution	75	75	75
Reagent III	250	250	250
Reagent IV	75	75	75
Mix thoroughly, water bath at 65℃ for 3 minutes.			
Reagent V	250	250	250
Reagent VI	250	250	250
Mix thoroughly, water bath at 65℃ for 3 minutes.			

Take out the EP tubes, colorized at 420 nm after cooling.

Note: Blank tube and standard tube only need to be measured once.

III. Calculation:

1 Serum:

$TG(mg/dL)= C\times(A_T- A_B)\div(A_S- A_B) \times100 =100 \times(A_T- A_B)\div(A_S- A_B)$

2 Tissure:

Protein concentration:

$TG(mg/mg\ prot)= C\times V\times(A_T- A_B)\div(A_S- A_B) \div(Cpr\times V)=(A_T- A_B)\div(A_S- A_B) \div Cpr$

Sample weight:

$TG(mg/g) = C\times V\times(A_T- A_B)\div(A_S- A_B) \div W=(A_T- A_B)\div(A_S- A_B) \div W$

3 Bacteria or cell:

$TG(U/10^4\ cell) = C\times(A_T- A_B)\div(A_S- A_B) \div D=(A_T- A_B)\div(A_S- A_B) \div D$

V: The volume of reagent1, 1mL;

C: Standard concentration, 1mg/ mL;

100:1dL=100 mL;

Cpr: Sample protein concentration (mg/mL);

W: Sample weight(g);

D: Density of bacteria or cell, 10^4 cell/mL.

Note:

1. There are volatile substances in the kit. Gloves and masks should be worn during the experiment. The reagent bottle cap should be closed in time after opening.

2. After the addition of Reagent II, it is necessary to repeatedly and violently vibrate, so that the triglyceride in the test solution can be fully extracted, and the oscillation amplitude, time, repeated times and waiting for stratification time should be consistent.

3. In order to ensure the repeatability of the test, the cooling time after each water bath should be unified.

4. If the OD value of the test tube is greater than 1, it is recommended to dilute the sample with Reagent I properly before testing, and multiply it by the corresponding dilution multiple during calculation.

Recent Products Citations:

[1] Wei Hu,Rui Wei,Liyue Wang,et al. Correlations of MMP-1, MMP-3,and MMP-12 with the degree of atherosclerosis, plaque stability and cardiovascular and cerebrovascular events. Experimental and Therapeutic Medicine. 2018;(IF1.448)

[2] Jieyong Xing,Yanshao Liu,Tao Chen. Correlations of chemokine CXCL16 and TNF-α with coronary atherosclerotic heart disease. Experimental and Therapeutic Medicine. November 2017;(IF1.448)

[3] Zhenbin Xu,Xizhuang Bai. Strontium ranelate-induced anti-adipocytic effects are involved in negative regulation of autophagy in rat bone marrow mesenchymal stem cells. International Orthopaedics. October 2018;(IF2.384)

[4] Chu X Y, Yang S Z, Zhu M Q, et al. Isorhapontigenin Improves Diabetes in Mice via Regulating the Activity and Stability of PPARγ in Adipocytes[J]. Journal of Agricultural and Food Chemistry, 2020, 68(13): 3976-3985.

[5] Li W, Li Y, Zhao Y, et al. The protective effects of aloperine against ox-LDL-induced endothelial dysfunction and inflammation in HUVECs[J]. Artificial Cells, Nanomedicine, and Biotechnology, 2020, 48(1): 107-115.

References:

[1] Fletcher M J. A colorimetric method for estimating serum triglycerides[J]. Clinica Chimica Acta, 1968, 22(3): 393-397.

[2] Hercules D M, Sheehan T L. Chemiluminescent determination of serum glycerol and triglycerides[J]. Analytical chemistry, 1978, 50(1): 22-25.

Related Products:

NA0733/NA0491	Free Cholestenone(FC) Content Assay Kit
NA0808/NA0566	Acetaldehyde Dehydrogenase(ALDH) Activity Assay Kit
NA0834/NA0592	Acetyl CoA carboxylase(ACC) Activity Assay Kit
NA0727/NA0485	Total Cholesterol(TC) Content Assay Kit

Technical Specifications:

The detection limit: 0.0559 mg/mL

Linear range: 0.0625-1.2 mg/mL