版本：A5　　修改日期：2024.12.11

Trizol(总 RNA 提取试剂)

产品简介：

Trizol 是一种新型的用于细胞或组织的总 RNA 提取试剂，Leagene Trizol 采用与 Invitrogen TRIzol 相似的原理和方法，其颜色、抽提的方法和步骤与后者完全相同。Leagene Trizol 含酚和异硫氰酸胍等物质，能迅速裂解细胞或组织并且灭活核酸酶，保持 RNA 的完整性。加入氯仿并离心后，溶液形成上清层为水相(无色)、中间层、下层为有机相(红色)；上清层用异丙醇沉淀回收总 RNA，中间层用乙醇沉淀回收 DNA，下层用异丙醇沉淀回收蛋白。

Leagene Trizol 适用于从各种组织或细胞中快速分离总 RNA，既可用于小量样品(50～100 mg 组织、5×10^6 细胞)，也可用于大量样品(>1g 组织/$>10^7$ 细胞)，提取的总 RNA 质量高，可用于 Northern blot、Dot blot、polyA 筛选、体外翻译、RNase 保护分析和分子克隆，该试剂具有以下特点：①适用范围广；②操作简单，整个过程 1 小时内完成；③纯度高；④污染少。该试剂仅用于科研领域，不适用于临床诊断或其他用途。

产品组成：

编号 / 名称	NR0002	Storage
Trizol Reagent	100ml	4℃ 避光
使用说明书	1 份	

自备材料：

1、试剂：无水乙醇、氯仿、异丙醇、DEPC 处理水等。

2、耗材： RNase 广泛存在于人的皮肤上和体液及环境中，RNase 是导致 RNA 降解的最主要物质，非常稳定。操作时应佩戴一次性口罩、手套、帽子。塑料制品、玻璃和金属物品、实验仪器等应清除 RNase，移液器吸头、EP 管等制品的 RNase free 处理尤为重要。

3、仪器：低温高速离心机、低温冰箱。

操作步骤(仅供参考)：

1、样品准备

⑴贴壁细胞：

①直接裂解：直接在培养瓶/皿中加入 Trizol 裂解细胞，每 $10cm^2$ 面积加 1ml Trizol，用移液器吹打混匀。

②胰蛋白酶消化：用无菌 PBS 洗涤细胞后，加入含有 0.05～0.25%胰蛋白酶的 PBS 处理细

胞，当细胞脱离容器壁后，加入含有血清的培养基终止反应，将细胞溶液转移至无 RNase 的离心管中，5000～6000g 离心 5 min，收集细胞沉淀，去除上清。收集细胞时一定要将细胞培养液去除干净，否则裂解不完全，降低 RNA 收获率。

⑵悬浮细胞：无需清洗细胞，直接 5000～6000 g 离心 5 min，收集细胞，每 5×10^6～10^7 动物、植物和酵母细胞或每 10^7 细菌细胞加入 1ml Trizol。

⑶组织：取新鲜动物或者植物组织或者-70℃冻存组织，50～100mg 组织在液氮中充分研磨或者加入 1ml Trizol 研磨或者用匀浆器匀浆处理。样品体积一般不超过 Trizol 体积的 10%，研磨要迅速，以 1min 为佳。

⑷血液：取 0.5～1 ml 新鲜或冻存的血液，12000g 离心 5 min，去除血浆，加入 1ml Trizol，充分振荡混匀。

2、 核酸分离：充分振荡混匀(可以置于低温/超低温冰箱冻存 5～10 min 后，充分振荡，反复 1～3 次)，将裂解样品或匀浆液室温放置 5～10 min，使核蛋白与核酸完全分离。

3、 样品分层：加入 0.2 ml 氯仿/1ml Trizol，剧烈振荡 15 s，室温放置 2～3 min，置于 4℃离心机，12000 g 离心 10～15 min；上层为水相，中间层和下层为有机相，RNA 在上层水相。

4、 沉淀 RNA：吸取上层水相(约 500μl)转移至无 RNase 的离心管中(不要吸取任何中间层物质,否则会有染色体 DNA 污染)，加入等体积异丙醇混匀(或者加入 1.2 倍体积 Leagene Trizol 专用 RNA 沉淀液，超低温冰箱放置 2～3hr,可以大大提高 RNA 回收率)，室温放置 15～20 min，12000 g 4℃离心 10 min，离心后管侧或管底形成胶状沉淀，弃上清。

5、 洗涤 RNA：加入 1ml DEPC 水配制的 75%乙醇/1ml Trizol 洗涤沉淀(或者加入 1ml Leagene Trizol 专用 RNA 洗涤液，可以大大提高 RNA 纯度)，室温放置 5～10 min，7500g 4℃离心 5 min，弃上清，室温干燥 5～10 min，不宜过分干燥，否则 RNA 难以溶解。

6、 溶解 RNA：加入 30～50ul RNase-free ddH2O 充分溶解 RNA，-70℃长期保存或直接用于后续试验。对于肝、胰腺、肾等组织中 RNase 含量高的样品，沉淀时用 100%去离子甲酰胺溶解。

分析与定量：

1、 测定样品在 260 nm 和 280 nm 的吸收值确定 RNA 的质量，按 1OD=40pg RNA 计算 RNA 的产率，$OD_{260/280}$ 在 1.8～2.0 视为抽提 RNA 纯度较好，浓度在 4μg/ml 以上的样品适于用分光光度计测定。

2、 进行甲醛变性琼脂糖电泳，确定 RNA 的完整性和污染情况。

3、 核酸分析仪测定 RNA 的质量和纯度。

注意事项：

1、样品保存：加入 Trizol 混匀后，样品可在-70℃放置 1～2 月；RNA 样品可以在 70%酒精中-70℃保存 2～4 周：如果需要长期保存，应置于超低温冰箱中保存。

2、Trizol 是强腐蚀性物质，污染皮肤或眼睛后，立即用清水或生理盐水冲洗，必要时寻求医生的帮助。

3、Trizol 可常温运输，建议保存 4℃保存。

4、为了您的安全和健康，请穿实验服并戴一次性手套操作。

5、试剂开封后请尽快使用，以防影响后续实验效果。

有效期：12 个月有效。常温运输，4℃保存。

常见问题分析：

常见问题	可能原因
$A_{260}/A_{280}<1.6$	抽提得到的 RNA 沉淀未完全溶解。
	水相中混有有机相，从而存在蛋白质和 DNA 污染。
	RNA 样品用水而不是 TE 溶解。低离子浓度和低 pH 条件下，A280 值会偏高。
	样品匀浆时加的 Trizol 试剂太少，RNA 与蛋白质、DNA 未能完全分离。
	匀浆后样品未在室温放置或者放置时间太短，RNA 与核蛋白未完全解离。
DNA 污染	样品中含组织溶剂(如乙醇等)或碱性溶液，致水相减少或 pH 升高。
	样品匀浆时加入的试剂体积太少。如果存在 DNA 污染，可用 DNA 清除剂去除。
	水相中混有有机相，从而存在蛋白质和 DNA 污染。
RNA 产量低	样品裂解或匀浆处理不彻底，RNA 没有被完全释放出来。
	得到的 RNA 沉淀未完全溶解。
	抽提的 RNA 中含有 RNase。
RNA 降解	组织或细胞不新鲜，样品没有及时被液氮冻存，导致组织或细胞中的 RNA 降解。
	溶液或离心管未经 RNase free 处理，RNase 的污染导致 RNA 被降解。
	细胞在胰蛋白酶消化时间过长，导致未加 Trizol 前 RNA 已经部分降解。
	电泳时使用的甲酰胺 pH 小于 3.5，导致 RNA 发生酸解。
蛋白和多糖污染	水相中混有有机相，从而带有蛋白质和 DNA。
	样品中蛋白、多糖含量高或样品量太大，细胞未裂解完全。

相关产品：

产品编号	产品名称
OR0001	pH 标准缓冲溶液(pH=4.00)
TC0713	葡萄糖检测试剂盒(GOD-POD 比色法)

文献引用：

1、 Xin mei Duan,Xu Yang,Nianlian Mou,et al.Reactive Oxygen Species-Triggered Carbon Dots-Based Biomimetic Nanotheranostic Agents for Atherosclerosis Management.ADVANCED FUNCTIONAL MATERIALS.July 2024.10.1002/adfm.202405629.(IF 18.5)
2、 Men gyue Wang,Xun Zhou,Siyi Zhou,et al.Mechanical force drives the initial mesenchymal-epithelial interaction during skin organoid development.Theranostics.May 2023.10.7150/thno.83217.(IF 12.4)
3、 Siy i Zhou,Zeming Li,Xinzhu Li,et al.Crosstalk between endothelial cells and dermal papilla entails hair regeneration and angiogenesis during aging.Journal of Advanced Research.May 2024.10.1016/j.jare.2024.05.006.(IF 11.4)
4、 Zhu xian Wang,Yi Hu,Yaqi Xue,et al.4′-OH as the Action Site of Lipids and MRP1 for Enhanced Transdermal Delivery of Flavonoids.ACS Applied Materials & Interfaces.March 2023.10.1021/acsami.2c18086.(IF 9.5)
5、 Jia lu Luan,Liang Wen,Yehua Bao,et al.Systemic toxicity of biodegradable polyglycolic acid microplastics on the gut-liver-brain axis in zebrafish.SCIENCE OF THE TOTAL ENVIRONMENT.October 2024.10.1016/j.scitotenv.2024.176898.(IF 8.2)
6、 Tia n Zhu,Chunxia Yao,Shuang Hong,et al.Multi-omics reveal toxicity mechanisms underpinning nanoplastic in redclaw crayfish (Cherax quadricarinatus).SCIENCE OF THE TOTAL ENVIRONMENT.August 2024.10.1016/j.scitotenv.2024.175680.(IF 8.2)
7、 Men gying Yang,Jinyu Qiu,Xin Zhao,et al.6-benzylaminopurine exposure induced development toxicity and behaviour alteration in zebrafish (Danio rerio).ENVIRONMENTAL POLLUTION.March 2021.10.1016/j.envpol.2021.116887.(IF 8.071)
8、 Qin g Tang,Xi Wang,Qichun Zhou,et al.Fuzheng Kang-Ai inhibits NSCLC cell proliferation via regulating hsa_circ_0048091/hsa-miR-378g/ARRDC3 pathway.PHYTOMEDICINE.April 2023.10.1016/j.phymed.2023.154819.(IF 7.9)
9、 Shu hui Zhang,Ping Mi,Jialu Luan,et al.Fluorene-9-bisphenol acts on the gut-brain axis by regulating oxytocin signaling to disturb social behaviors in zebrafish.ENVIRONMENTAL RESEARCH.May 2024.10.1016/j.envres.2024.119169.(IF 7.7)
10、Xu Zhang,Jian Li,Jin Chen,et al.Enhanced bone regeneration via PHA scaffolds coated with polydopamine-captured BMP2.Journal of Materials Chemistry B.July 2022.10.1039/D2TB01122K.(IF 7.571)
11、Lil i Fan,Xueying Zhang,Yihai Huang,et al.Homoplantaginin attenuates high glucose-induced vascular endothelial cell apoptosis through promoting autophagy via the AMPK/TFEB pathway.PHYTOTHERAPY RESEARCH.March 2023.10.1002/ptr.7797.(IF 7.2)
12、Shu hui Zhang,Zhenhua Fu,Yixin Xu,et al.The masculinization steroid milieu caused by fluorene-9-bisphenol disrupts sex-typical courtship behavior in female zebrafish (Danio rerio).ECOTOXICOLOGY AND ENVIRONMENTAL SAFETY.October 2022.10.1016/j.ecoenv.2022.114174.(IF 7.129)
13、Yi- xin Xu,Shu-hui Zhang,Shao-zhi Zhang,et al.Exposure of zebrafish embryos to sodium propionate disrupts circadian behavior and glucose metabolism-related development.ECOTOXICOLOGY AND ENVIRONMENTAL SAFETY.June 2022.10.1016/j.ecoenv.2022.113791.(IF 7.129)
14、Shu hui Zhang,Xiang Zuo,Jialu Luan,et al.The deleterious effects and potential therapeutic strategy of fluorene-9-bisphenol on circadian activity and liver diseases in zebrafish and mice.Journal of Environmental Sciences.September 2023.10.1016/j.jes.2023.09.014.(IF 6.9)
15、Jia lu Luan,Shuhui Zhang,Yixin Xu,et al.Effects of microplastic exposure on the early developmental period and circadian rhythm of zebrafish (Danio rerio): A comparative study of polylactic acid and polyglycolic acid.ECOTOXICOLOGY AND ENVIRONMENTAL SAFETY.May 2023.10.1016/j.ecoenv.2023.114994. (IF 6.8)
16、Shu hui Zhang,Yixin Xu,Shaozhi Zhang,et al.Fluorene-9-bisphenol exposure decreases locomotor activity and induces lipid-metabolism disorders by impairing fatty acid oxidation in zebrafish.LIFE SCIENCES.February 2022.10.1016/j.lfs.2022.120379.(IF 6.78)
17、Men gying Yang,Jiaxing Huang,Shuhui Zhang,et al.Melatonin mitigated circadian disruption and cardiovascular toxicity caused by 6-benzylaminopurine exposure in zebrafish.ECOTOXICOLOGY AND ENVIRONMENTAL SAFETY.July 2021.10.1016/j.ecoenv.2021.112555.(IF 6.291)
18、Men gying Yang,Jialu Luan,Yixin Xu,et al.Cardiotoxicity of Zebrafish Induced by 6-Benzylaminopurine Exposure and Its Mechanism.INTERNATIONAL JOURNAL OF MOLECULAR SCIENCES.July 2022.10.3390/ijms23158438.(IF 6.208)
19、Xin ping Guo,Ziang Qian,Su Jiang,et al.Assessing the ecotoxicity of florfenicol exposure at environmental levels: A case study of histology, apoptosis and microbiota in hepatopancreas of Eriocheir sinensis.ECOT

OXICOLOGY AND ENVIRONMENTAL SAFETY.February 2024.10.1016/j.ecoenv.2024.116092.(IF 6.2)

20、Wan Huan,You Ting,et al.circ_0003204 Regulates Cell Growth, Oxidative Stress, and Inflammation in ox-LDL-Induced Vascular Endothelial Cells via Regulating miR-942-5p/HDAC9 Axis.Frontiers in Cardiovascular Medicine.April 2021.10.3389/fcvm.2021.646832.(IF 6.05)

21、Kai hui Xie,Zunqiang Yan,Qiaoli Yang,et al.lnc001776 Affects CPB2 Toxin-Induced Excessive Injury of Porcine Intestinal Epithelial Cells via Activating JNK/NF-kB Pathway through ssc-let-7i-5p/IL-6 Axis.Cells.March 2023.10.3390/cells12071036.(IF 6)

22、Liu Mengqian,Wei Xuerong,Zheng Zijun,et al.AMPK activation eliminates senescent cells in diabetic wound by inducing NCOA4 mediated ferritinophagy.MOLECULAR MEDICINE.May 2024.10.1186/s10020-024-00825-8.(IF 6)

23、Pin g Mi,Qian Gao,Ze-Yang Feng,et al.Melatonin attenuates 17β-trenbolone induced insomnia-like phenotype and movement deficiency in zebrafish.CHEMOSPHERE.April 2020.10.1016/j.chemosphere.2020.126762.(IF 5.778)

24、Gui ying Sun,Hua Ye,Huijuan Liu,et al.ZPR1 is an immunodiagnostic biomarker and promotes tumor progression in esophageal squamous cell carcinoma.CANCER SCIENCE.November 2023.10.1111/cas.16004.(IF 5.7)

25、Wan g Ting,Li Chaopeng,Shi Min,et al.Circular RNA circZNF532 facilitates angiogenesis and inflammation in diabetic retinopathy via regulating miR-1243/CARM1 axis.Diabetology & Metabolic Syndrome.-01 2022.10.1186/s13098-022-00787-z.(IF 5.395)

26、Hua ng Mei,Guo Tian,Meng Yan,et al.Comprehensive analysis of the prognosis and immune effect of the oncogenic protein Four Jointed Box 1.Frontiers in Oncology.May 2023.10.3389/fonc.2023.1170482.(IF 4.7)

27、Lia ng Yi,Lei Ouyang,Shuang Wang,et al.Long noncoding RNA PTPRG-AS1 acts as a microRNA-194-3p sponge to regulate radiosensitivity and metastasis of nasopharyngeal carcinoma cells via PRC1.JOURNAL OF CELLULAR PHYSIOLOGY.April 2019.10.1002/jcp.28547.(IF 4.522)

28、Xin Shen,Hai Shi,Xinli Chen,et al.Esculetin Alleviates Inflammation, Oxidative Stress and Apoptosis in Intestinal Ischemia/Reperfusion Injury via Targeting SIRT3/AMPK/mTOR Signaling and Regulating Autophagy.Journal of Inflammation Research.August 2023.10.2147/JIR.S413941.(IF 4.5)

29、Na Wang,Yunfeng Guo,Liqin Song,et al.Circular RNA intraflagellar transport 80 facilitates endometrial cancer progression through modulating miR-545-3p/FAM98A signaling.Journal of Gynecologic Oncology.October 2021.10.3802/jgo.2022.33.e2.(IF 4.401)

30、Qia n Tian,Huo Bengang,Deng Xiaorong,et al.Decreased TAX1BP1 participates in systemic lupus erythematosus by regulating monocyte/macrophage function.INTERNATIONAL IMMUNOLOGY.July 2023.10.1093/intimm/dxad027.(IF 4.4)

31、Yu Wang,Xiangyun Chen,Qiduan Chen,et al.Expression of human dCTP pyrophosphatase 1 (DCTPP1) and its association with cisplatin resistance characteristics in ovarian cancer.JOURNAL OF CELLULAR AND MOLECULAR MEDICINE.April 2024.10.1111/jcmm.18371.(IF 4.3)

32、Ma Junfu,Meng Qingliang,Zhan Junping,et al.Paeoniflorin Suppresses Rheumatoid Arthritis Development via Modulating the Circ-FAM120A/miR-671-5p/MDM4 Axis.INFLAMMATION.August 2021.10.1007/s10753-021-01504-0.(IF 4.092)

33、Zhe ng Juyan,Zhang Wei,Kang PeiPei,et al.Midazolam Ameliorates Impairment of the Blood–Brain Barrier (BBB) Against LPS.NEUROTOXICITY RESEARCH.April 2022.10.1007/s12640-022-00508-4.(IF 3.978)

34、Wu Yao,Bai Yu,Lu Yan,et al.Transcriptome sequencing and network pharmacology-based approach to reveal the effect and mechanism of Ji Chuan Jian against Parkinson's disease.BMC Complementary Medicine and Therapies.June 2023.10.1186/s12906-023-03999-6.(IF 3.9)

35、Lia ng Wen,Xiaoting Man,Jialu Luan,et al.Early-life exposure to five biodegradable plastics impairs eye development and visually-mediated behavior through disturbing hypothalamus-pituitary-thyroid (HPT) axis in zebrafish larvae.COMPARATIVE BIOCHEMISTRY AND PHYSIOLOGY C-TOXICOLOGY & PHARMACOLOGY.July 2024.10.1016/j.cbpc.2024.109981.(IF 3.9)

36、Pen g Chu,Sheng Luo,Haozhe Wang,et al.Interactive effects of temperature and salinity on the apoptosis, antioxidant enzymes, and MAPK signaling pathway of juvenile pufferfish (Takifugu fasciatus).Aquaculture Reports.February 2023.10.1016/j.aqrep.2023.101483.(IF 3.7)

37、Cha ng Liu,Jin-Ou Wang,Wen-Yang Zhou,et al.Long non-coding RNA LINC01207 silencing suppresses AGR2 expression to facilitate autophagy and apoptosis of pancreatic cancer cells by sponging miR-143-5p.MOLECULAR AND CELLULAR ENDOCRINOLOGY.April 2019.10.1016/j.mce.2019.04.004.(IF 3.693)

38、Wan g Shiliang,Li Wenlu,Yang Long,et al.CircPVT1 facilitates the progression of oral squamous cell carcinoma by regulating miR-143-3p/SLC7A11 axis through MAPK signaling pathway.FUNCTIONAL & INTEGR

ATIVE GENOMICS.May 2022.10.1007/s10142-022-00865-5.(IF 3.674)
39、Lil i Zhu,Dongwu Shi,Jianghong Cao,et al.LncRNA CASC2 Alleviates Sepsis-induced Acute Lung Injury by Regulating the miR-152-3p/PDK4 Axis.IMMUNOLOGICAL INVESTIGATIONS.June 2021.10.1080/08820139.2021.1928693.(IF 3.657)
40、Wan g Xiaoyu,Liu Lili,Zhang Lei,et al.Circ_0057583 facilitates brain microvascular endothelial cell injury th rough modulating miR-204-5p/NR4A1 axis.METABOLIC BRAIN DISEASE.November 2021.10.1007/s11011-021-00866-6.(IF 3.584)
41、Yan g Ling,Wang Lijuan,Wang Juan,et al.Long non-coding RNA Gm11974 aggravates oxygen–glucose de privation-induced injury via miR-122-5p/SEMA3A axis in ischaemic stroke.METABOLIC BRAIN DISEASE.August 2021.10.1007/s11011-021-00792-7.(IF 3.584)
42、Kai hui Xie,Qiaoli Yang,Zunqiang Yan,et al.Identification of a Novel lncRNA LNC_001186 and Its Effects o n CPB2 Toxin-Induced Apoptosis of IPEC-J2 Cells.Genes.May 2023.10.3390/genes14051047.(IF 3.5)
43、Jie Gao,Chengzhi Ding,Junhui Zhou,et al.Propofol suppresses lung cancer tumorigenesis by modulating t he circ-ERBB2/miR-7-5p/FOXM1 axis.Thoracic Cancer.-01 2021.10.1111/1759-7714.13856.(IF 3.5)
44、Min gming Gu,Kun Liu,Hui Xiong,et al.MiR-130a-3p inhibits endothelial inflammation by regulating the exp ression of MAPK8 in endothelial cells.Heliyon.-01 2024.10.1016/j.heliyon.2024.e24541.(IF 3.4)
45、Che n Feng,Jiang Ruilai,et al.Circular RNA circ_0002984 Facilitates the Proliferation and Migration of Ox-LDL-Induced Vascular Smooth Muscle Cells via the Let-7a-5p/KLF5 Pathway.Cardiovascular Toxicology.August 2024.10.1007/s12012-024-09911-z.(IF 3.4)
46、Jiu mei Cai,Xiaoping Wang,Wei Liao,et al.Silencing lncRNA 93358 Inhibits the Apoptosis of Myocardial C ells in Myocardial Infarction Rats by Inducing the Expression of SLC8A1.Biomed Research International.July 2022.10.1155/2022/1138709.(IF 3.246)
47、Min jie He,Liang Tu,Ruo Shu,et al.Long Noncoding RNA CASC2 Facilitated Wound Healing through miR NA-155/HIF-1α in Diabetic Foot Ulcers.Contrast Media & Molecular Imaging.June 2022.10.1155/2022/6291497.(IF 3.009)
48、Jia Song,Jia Li,Xiaochen Pei,et al.Identification of cuproptosis-realated key genes and pathways in Parkinson's disease via bioinformatics analysis.PLoS One.April 2024.10.1371/journal.pone.0299898.(IF 2.9)
49、Jia n-Hua Liu,Chen Li,Liang Cao,et al.Exosomal miR-132-3p from mesenchymal stem cells alleviated LPS -induced acute lung injury by repressing TRAF6.AUTOIMMUNITY.September 2021.10.1080/08916934.2021.1966768.(IF 2.815)
50、Lin Zenghai,et al.Circ_0004585 Facilitates Tumorigenesis of Colorectal Cancer Via Modulating the miR-338-3p/ZFX Axis and Activating the MEK/ERK Pathway.Cellular and Molecular Bioengineering.-01 2023.10.1007/s12195-022-00756-6.(IF 2.8)

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