读书报告

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RESEARCH ARTICLE

Grape seed proanthocyanidins repress the hepatic lipid regulators miR-33 and miR-122 in rats

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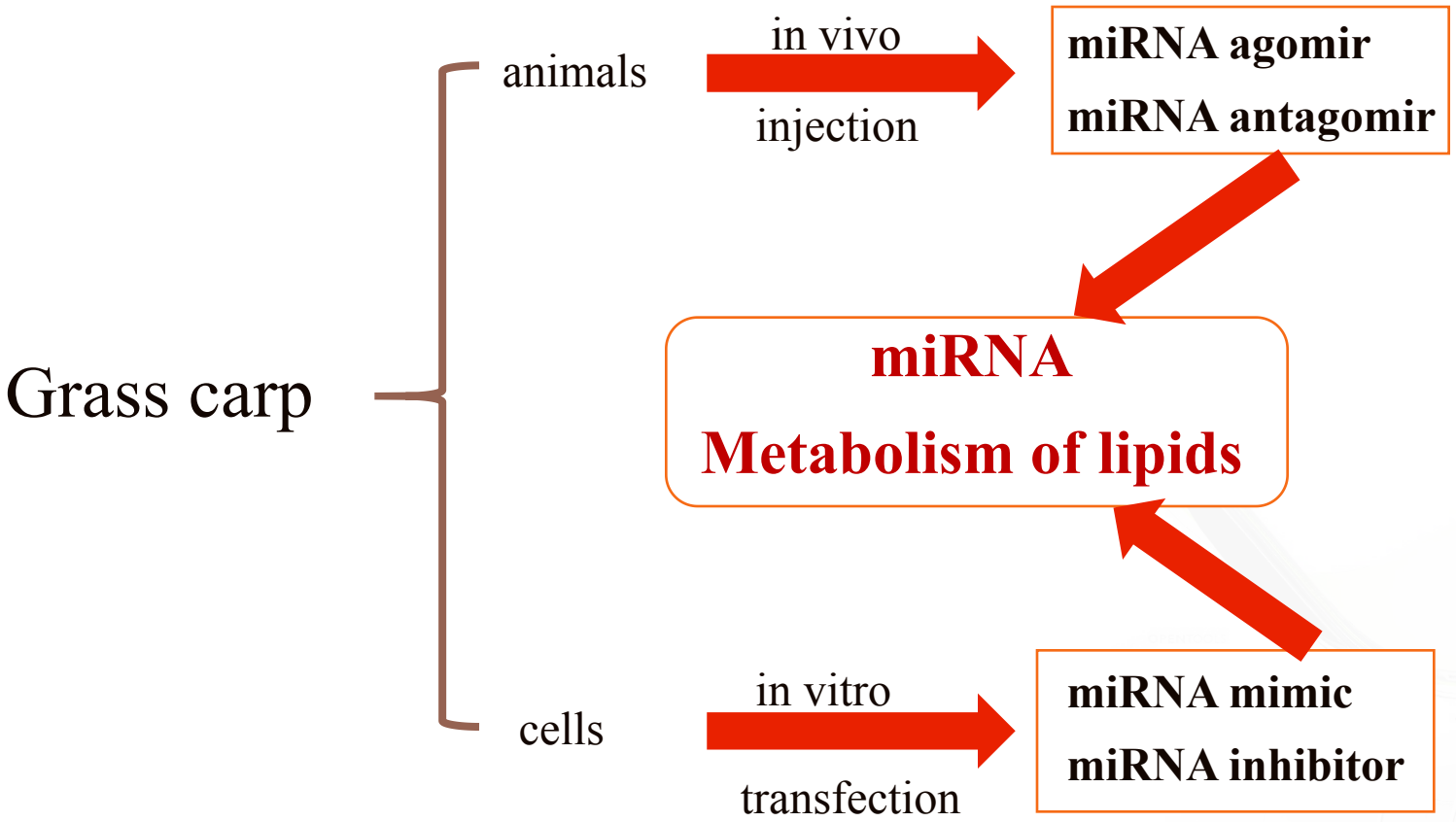
Grass carp
animals
in vivo
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Metabolism of lipids
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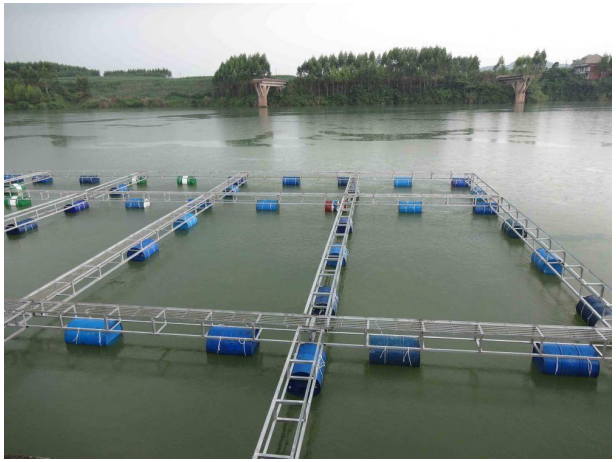
in vitro
transfection
miRNA mimic
miRNA inhibitor

Aquaculture



feed additive

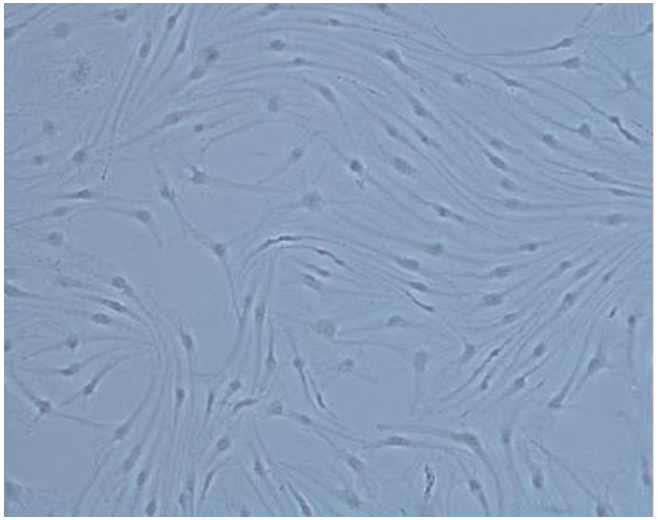
papers

Chinese herbal medicine

industrial production

High-lipid diet

(fish oil & soybean oil)



oleic acid

什么是葡萄籽提取物？

葡萄籽提取物是从葡萄籽中提取的一种人体内不能合成的新型高效天然抗氧化剂物质。它是目前自然界中发现的抗氧化、清除自由基能力最强的物质，其抗氧化活性为维生素E的50倍、维生素C的20倍，它能有效清除人体内多余的自由基，具有超强的延缓衰老和增强免疫力的作用。抗氧化、抗过敏、抗疲劳增强体质、改善亚健康状态延缓衰老、改善烦躁易怒、头昏乏力、记忆力减退等症状。

摘选自百度百科 “葡萄籽提取物”

什么是葡萄籽提取物？

葡萄籽中含有多酚类物质(GSP)，主要有儿茶素类和**原花青素类**。儿茶素类化合物包括儿茶素、表儿茶素及其没食子酸酯，是葡萄籽中主要的单聚体，也是原花青素寡聚体和多聚体的构成单位。约含有10 %~15 %的葡萄籽油，其主要成分为亚油酸、亚麻酸等多种不饱和脂肪酸和甾醇等以及多羟基芪类(PHS)如白藜芦醇等。

Antioxidant

什么是原花青素？

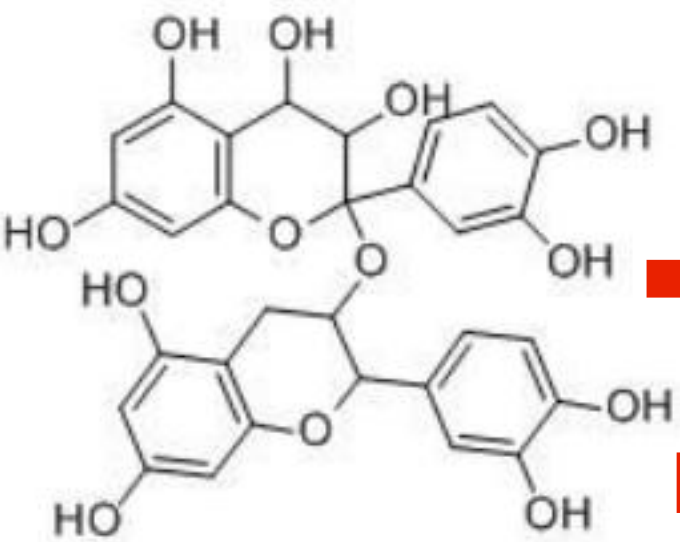
原花青素是Oligomeric Proantho Cyanidins（OPC）的中文学名，是一种有着特殊分子结构的生物类黄酮，是目前国际上公认的清除人体内自由基最有效的天然抗氧化剂。一般为红棕色粉末，气微、味涩，溶于水和大多有机溶剂。 一般为葡萄籽提取物或法国海岸松树皮提取物。

20世纪70年代，研究者又发现了获得OPC的另一个更好的资源——葡萄籽。**用葡萄籽提取的OPC含量高达95%，并且他还用葡萄籽中的OPC系统地做了一系列的实验，如生物利用度实验、毒性实验、三致实验(致畸、致癌、致突变)等，发现葡萄籽OPC是迄今为止最好的opc来源。**

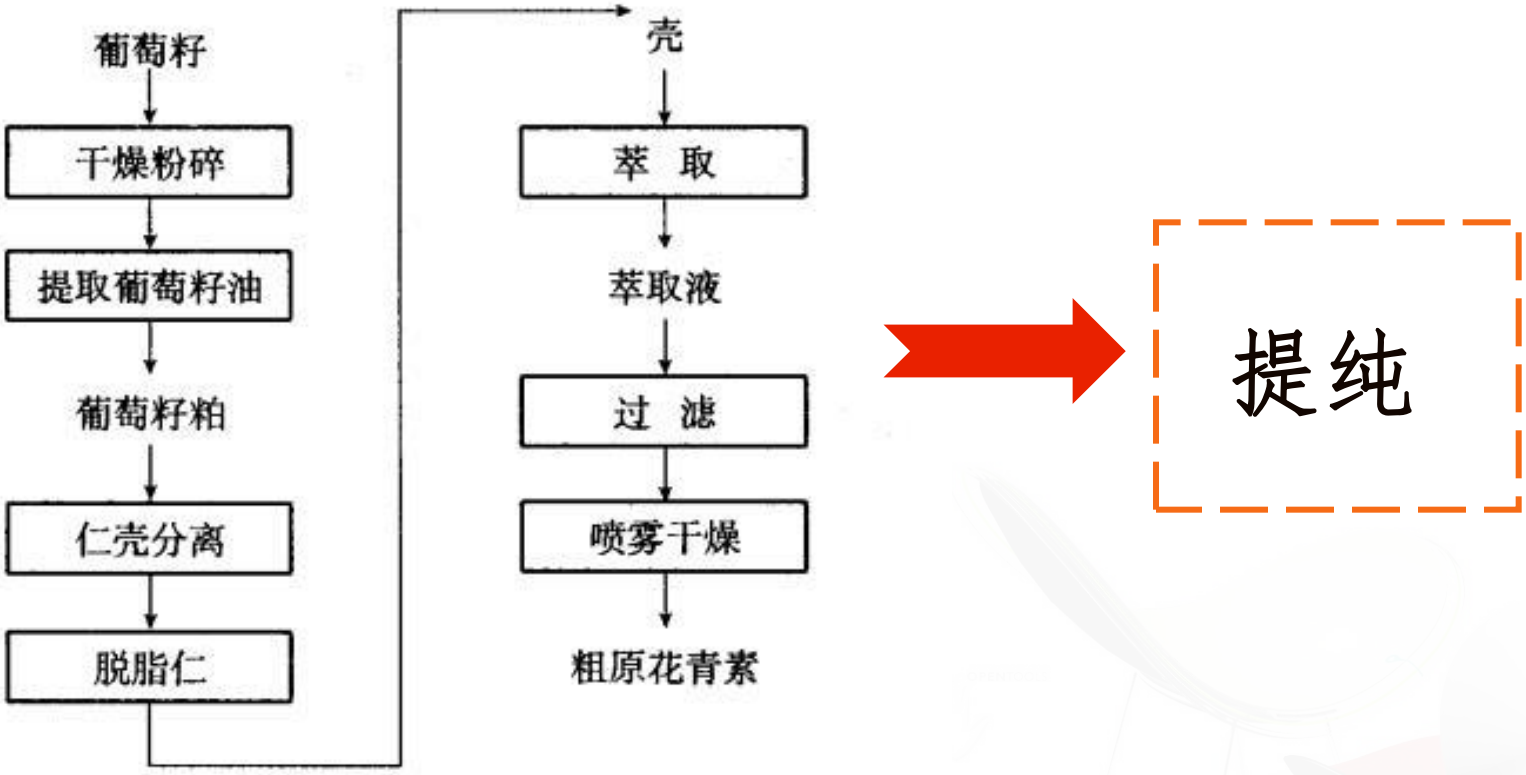
什么是原花青素？

原花青素（葡萄籽提取物）是一种新型高效抗氧化剂，是目前为止所发现的最强效的自由基清除剂，**具有非常强的体内活性**。实验证明，OPC的抗自由基氧化能力是维生素E的50倍，维生素C的20倍，并吸收迅速完全，**口服20分钟即可达到最高血液浓度**，代谢半衰期达7小时之久。

原花青素的作用是怎样的？



原花青素的制作工艺是怎样的？



来源于《植提网》“ 葡萄籽提取物原花青素生产的关键技术”

http://www.pewiki.net/opc-sheng-chan/?replytocom=2352

原花青素饲料添加剂在动物生产中的应用 - 添... 中国饲料行业信息网

2016年3月2日 - 原花青素饲料添加剂在动物生产中的应用 李海利1 周松涛1 巩耀进1 崔嘉3 谷子林1,2 (1.河北农业大学动物科技学院,保定071001;2.河北省山区研究所...

www.feedtrade.com.cn/a... - 百度快照 - 85%好评

原花青素的生物学功能及其在饲料上及应用 - 豆丁网

2015年7月9日 - 原花青素的生物学功能及其在饲料上及应用,原花青素,原花青素的功效与作用,原花青素清基胶囊,原花青素多少钱一盒,原花青素胶囊价格,全民健原花青素,原...

www.docin.com/p-121501... - 百度快照 - 2330条评价

原花青素饲料添加剂在动物生产中的应用_百度学术

李海利，周松涛，巩耀进 - "京津冀一体化畜牧兽医科技创新研讨会暨"瑞普杯"新思想、新方法、新观点论坛" - 2014

原花青素是天然植物多酚,具有抗氧化、细胞保护和免疫调节等营养和药理作用等优点,在食品、医药和化妆品等领域得到了广泛的应用。本文就原花青素的生物特性、生理功能...

xueshu.baidu.com

原花青素饲料添加剂对家兔的营养和抗氧化作用_百度学术

李海利，周松涛，巩耀进 - 中国畜牧兽医学会学术年会 - 2014

原花青素(Procyanidins,PC)又名前青花素,在营养学上被归为黄酮类化合物,是由不同数量的儿茶素与表儿茶素结合而成,形成二聚体、三聚体直至十聚体。原花青素广泛存在于...

xueshu.baidu.com

原花青素的生物学功能及其在饲料上的应用_百度学术

何沙沙，文利新 - 中国畜牧兽医学会家畜内科学分会代表大会暨学术研讨会 - 2011

原花青素属于生物类黄酮家族,具有抗氧化、预防心血管疾病和癌症等多种生物学功能。本文综述了原花青素的各种生物学功能及其应用价值。

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饲料中添加葡萄籽提取物对罗非鱼生长性能的影响

来自万方

作者 卢俊姣，翟少伟，史庆超，李剑，孙秀文

摘要 为研究饲料中添加葡萄籽提取物对罗非鱼生长性能的影响，选取平均体重为(8.25±0.07)g的罗非鱼300尾，随机分为5个处理组，分别投喂添加0(对照组)、200、400、600和800mg/kg葡萄籽提取物的试验饲料；每个处理组4个重复，每个重复15尾鱼，试验期为7周.结果为：与对照组相比，添加葡萄籽提取物各处理组罗非鱼的增重率和特定增长率显著提高(P <0.05)，饲料系数显著降低(P <0.05)，各添加组间无显著差异(P >0.05).随葡萄籽提取物添加水平的增加，蛋白质效率先上升后下降，除800mg/kg添加组外的其他各添加组均显著高于对照组(P <0.05)；而各处理组间摄食率和存活率无显著差(P >0.05).表明，饲料中添加葡萄籽提取物可有效提高罗非鱼的生长性能.经计算分析，以增重率和饲料系数为评价指标，葡萄籽提取物最适添加量为450mg/kg. ▲ 收起

出版源 世界华人鱼虾营养学术研讨会, 2013

关键词 葡萄籽提取物 / 罗非鱼 / 生长性能

葡萄籽等植物提取物对湘云金鲫生长性能及肌肉成分的影响

黄光中，罗世民，曾宪文 - 《水产科学》 - 2012 - 被引量: 2

摘　要: 21～25℃水温下,经30d的饲养试验,探讨葡萄籽等植物提取物对湘云金鲫生长和鱼肉品质 的影响。试验结果表明,在基础饲料中添加10g/kg葡萄籽等植物提取物能显...

全部来源：万方 / 维普 / 知网 / airitilibrary.com

葡萄籽等植物提取物与人工水草在黄鳝养殖上的应用

黄光中 - 《湖南农业大学》 - 2011 - 被引量: 2

本研究以黄鳝为研究对象,探讨葡萄籽等植物提取物在黄鳝养殖中的作用。在黄鳝饲料中添加0%、2%、4%三种剂量植物提取物进行饲养,经过102天饲养,测定黄鳝增重率、饲料...

全部来源：知网

葡萄籽等植物提取物对黄鳝促生长机制的研究

黄光中，胡辉，罗世民，... - 《当代水产》 - 2012 - 被引量: 0

正植物提取物以其天然性、无耐 药性及多功能性而渐受青睐。植物提取物添加剂既能防治水产动物病害,减少抗生素和化学药物的使用,又可以提高养殖动物消化酶活性,调节...

全部来源：知网 / 维普 / 万方

Introduction

grape seed proanthocyanidin extract

The most abundant polyphenols in the human diet are the proanthocyanidins, a subclass of flavonoids . These compounds are mainly present in apples, grapes, nuts, red wine, tea, and cocoa . Proanthocyanidins improve human health with cardioprotectant , antigenotoxic , antiinflammatory , antioxidant , and anticarcinogenic activities.

grape seed proanthocyanidin extract

A grape seed proanthocyanidin extract (GSPE) was shown to reduce plasma triglycerides (TGs) levels, apo B and LDL cholesterol, as well as to increase the percentage of HDL cholesterol in healthy rats given an acute oral dose of GSPE . **The hypolipidemic effects of GSPE were even more obvious in a lipid tolerance test model** . Additionally, chronic treatment with GSPE corrects the dyslipidemia associated with dietary obesity in rats .

Several mechanisms by which GSPE induces hypolipidemia have already been described . For example,GSPE activates genes that control fatty acid oxidation and represses genes that control lipogenesis and VLDL assembly in the liver , thus inducing hypolipidemia.

However, it is becoming clear that microRNAs (miRNAs) play key roles in the regulation of genes involved in lipid metabolism in the liver, and the effects of GSPE on these miRNAs is unknown .

miRNA

miRNAs play important regulatory roles in a variety of biological processes. Specifically, several miRNAs have been correlated with obesity and metabolic syndrome and are proposed to regulate glucose metabolism , adipocyte differentiation and adipogenesis , and lipid metabolism . **Two of the best-studied miRNAs involved in the regulation of lipid metabolism are miR-122 and miR-33 .**

miR-122

miR-122 is liver specific and represents 70% of all miRNA expression in liver . Thedysregulation of this miRNA has been associated with the dysregulation of genes with key roles in the control of liver lipid metabolism. miR-122 regulates several genes that control fatty acid and TG biosynthesis, such as fatty acid synthase (Fas), acetyl-CoA carboxylase 1, acetyl-CoA carboxylase 2,stearoyl-CoA desaturase 1, diacylglycerol O-acyltransferase 2, ATP citrate lyase, and sterol regulatory element-binding protein 1c, as well as genes that regulate fatty acid β-oxidation,such as carnitine palmitoyltransferase 1α (CPT1α)

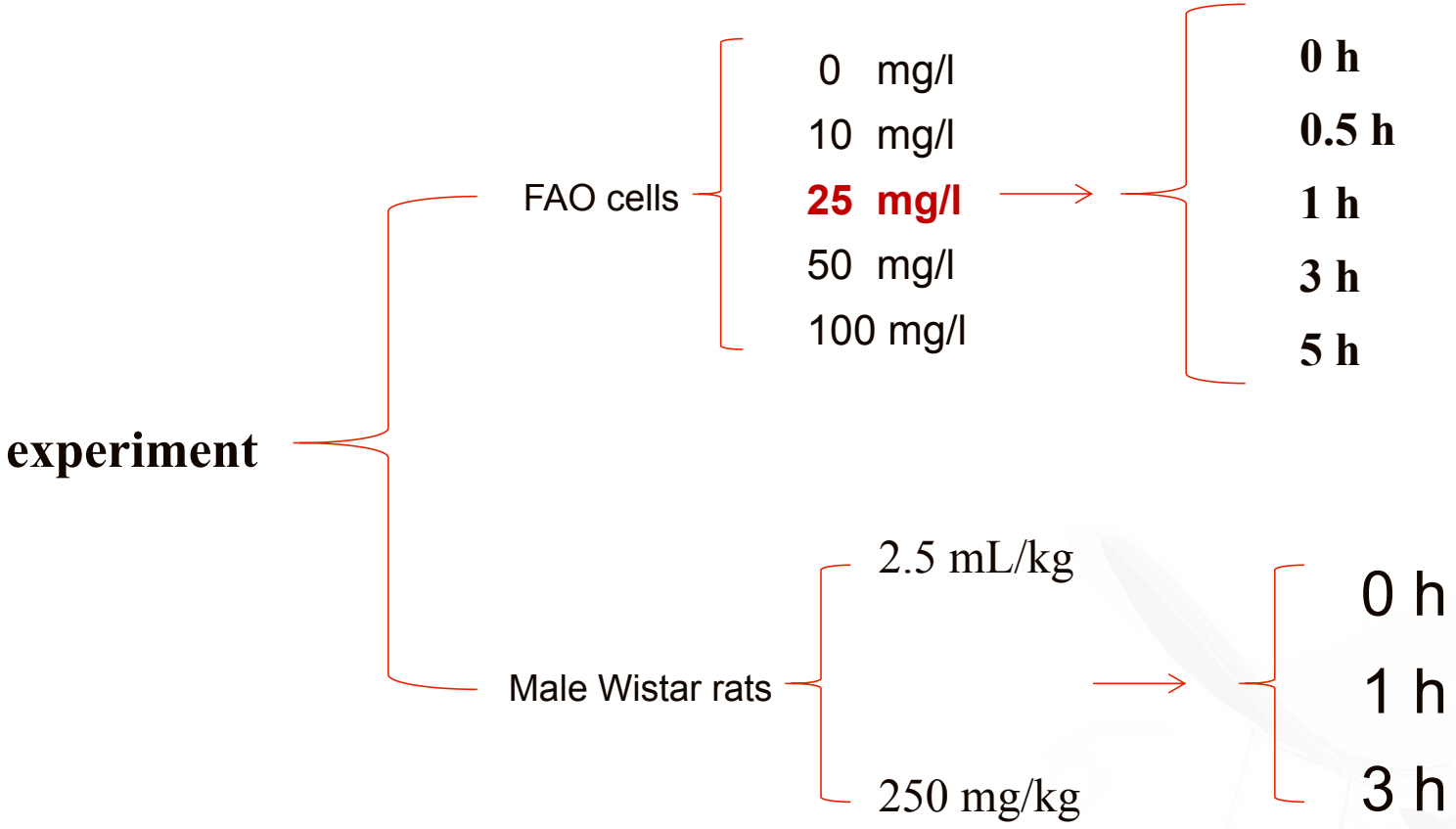
mir-122 microRNA precursor

miR-33

A second miRNA, **miR-33**, plays an important role in the regulation of cholesterol homeostasis, regulating the ATP-binding cassette transporters (ABC transporters),Abca1 and ABCG1, in addition to its role in fatty acid β-oxidation, where it regulates carnitine O-octanoyltransferase (CROT), hydroxyacyl-CoA dehydrogenase/3-ketoacyl-CoA thiolase/enoyl-CoA hydratase β-subunit (HADHB), and CPT1α .

Recently, it has been reported that dietary polyphenols such as curcumin , resveratrol , epigallocatechin gallate , ellagitannin , isoflavones , and proanthocyanidins from grape seed and cocoa modulate miRNA expression. Thus, we hypothesized that miRNAs could mediate the hypolipidemic effects of proanthocyanidins. Here, we test this hypothesis by examining the effects of GSPE on miR-122 and miR-33 levels in hepatic cells, using both in vivo and in vitro models.

experimental design



Materials and methods

Cells and cell culture

FAO cells, a rat hepatoma cell line (ECACC, code 85061112),

10% fetal bovine serum (BioWhittaker, Cologne, Germany) in a 95% air, 5% CO2 atmosphere at 37℃.

FAO 细胞是小鼠的肝癌细胞.因为它属于和原代肝细胞极为相似的一类细胞系,所以常被用来在体外进行肝细胞的研究.

The European Collection of Authenticated Cell Cultures (ECACC)

At 15 h before GSPE treatment, the media was replaced with serum-depleted media supplemented with 100 μM oleic acid and 40 μM BSA (bovine serum albumin, fatty acid free, Sigma-Aldrich). **FAO cells were treated with 10, 25, 50, or 100 mg GSPE per liter of media to select the working dose.** For kinetic experiments, cells were treated with 25 mg GSPE per liter of media. GSPE was dissolved in ethanol and added to the culture media; the final concentration of ethanol in the media was 0.05%, a nontoxic percentage. miRNAs and mRNAs were extracted at 0, 0.5, 1, 3, and 5 h after GSPE treatment.

Animals and experimental design

Male Wistar rats weighing 225 g

Animals were housed in animal quarters at 22℃ with a 12 h light/dark cycle (light from 8:00 to 20:00 h) and fed ad libitum with a standard chow diet (Panlab, Barcelona, Spain).

At 9 a.m. on the day of the experiment, the rats (five animals per group) were orally gavaged with lard oil (2.5 mL/kg body weight) (control group) or GSPE (250 mg/kg body weight) dissolved in lard oil (GSPE group).

At 0, 1, or 3 h after treatment, the rats were sedated using a combination of ketamine (70 mg ketamine/kg body weight) and xylazine (5 mg xylazine/kg body weight). After anesthetization, the rats were exsanguinated from the abdominal aorta. Blood was collected using heparin as an anticoagulant. **Plasma was obtained by centrifugation (1500 g, 15 min, 4℃) and stored at –80℃ until analysis.**The liver was excised, frozen immediately in liquid nitrogen, and stored at –80℃ until RNA and lipid extraction.

Results

GSPE decreased TG and cholesterol levels in plasma and liver

Table 3. Triglyceride and cholesterol levels in the plasma and livers of rats fed lard oil with or without proanthocyanidins (grape seed proanthocyanidin extract (GSPE))

	Basal (0 h)	Lard (3 h)	Lard + GSPE (3 h)
Plasma triglycerides (g/100 mL plasma)	71.4 ± 6.0	123.1 ± 6.6	73.4 ± 12.4[a)]
Plasma cholesterol (g/100 mL plasma)	18.5 ± 2.5	31.6 ± 6.1	26.1 ± 3.7
Total liver lipids (g/100 g liver)	6.7 ± 0.5	7.1 ± 0.5	5.5 ± 0.3[b)]
Liver triglycerides (g/100 g liver)	1.7 ± 0.2	2.4 ± 0.2	1.45 ± 0.33[a)]
Liver cholesterol (g/100 g liver)	0.9 ± 0.1	1.3 ± 0.1	0.9 ± 0.01[a)]

Rats fasted for 14 h were orally administered lard oil (2.5 mL/kg) with or without GSPE (250 mg/kg). Lipids were quantified before treatment (0 h) and at 3 h after GSPE administration. The values are the means of five animals per group.
a) Significant difference between the lard group and the lard + GSPE group.
b) Significant difference between the basal group and the lard + GSPE group ($p < 0.05$; Student's t test).

Results

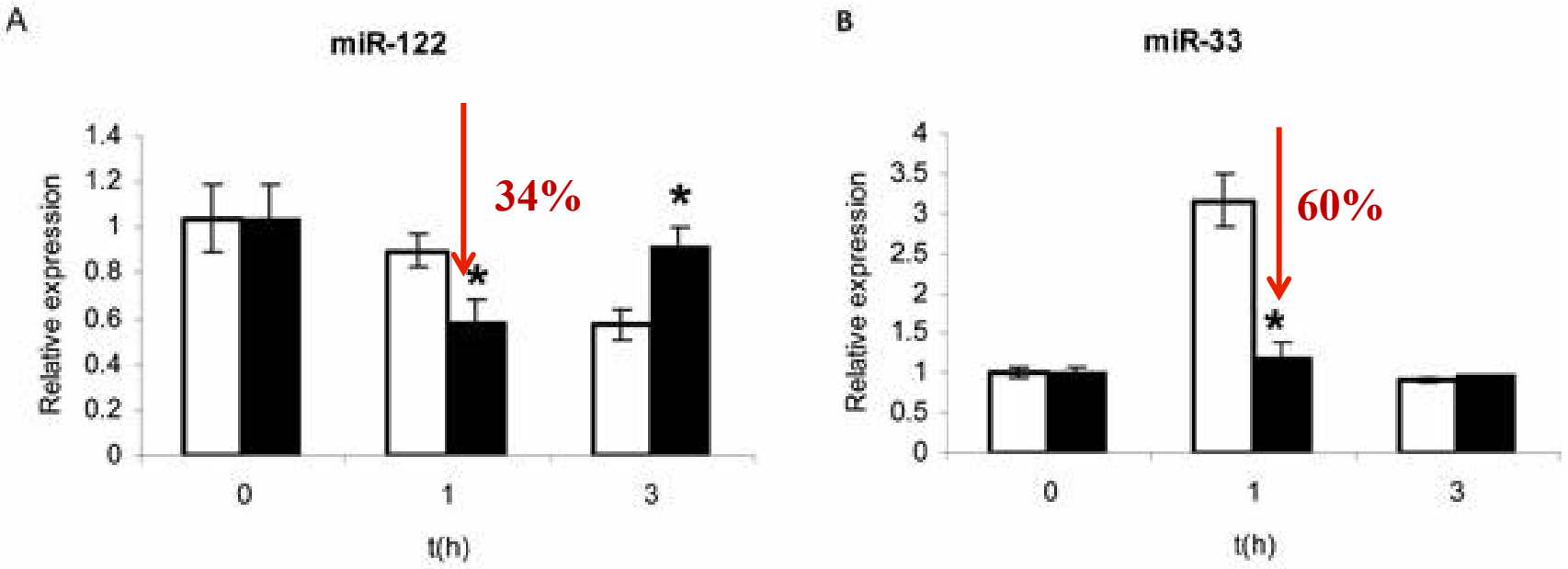
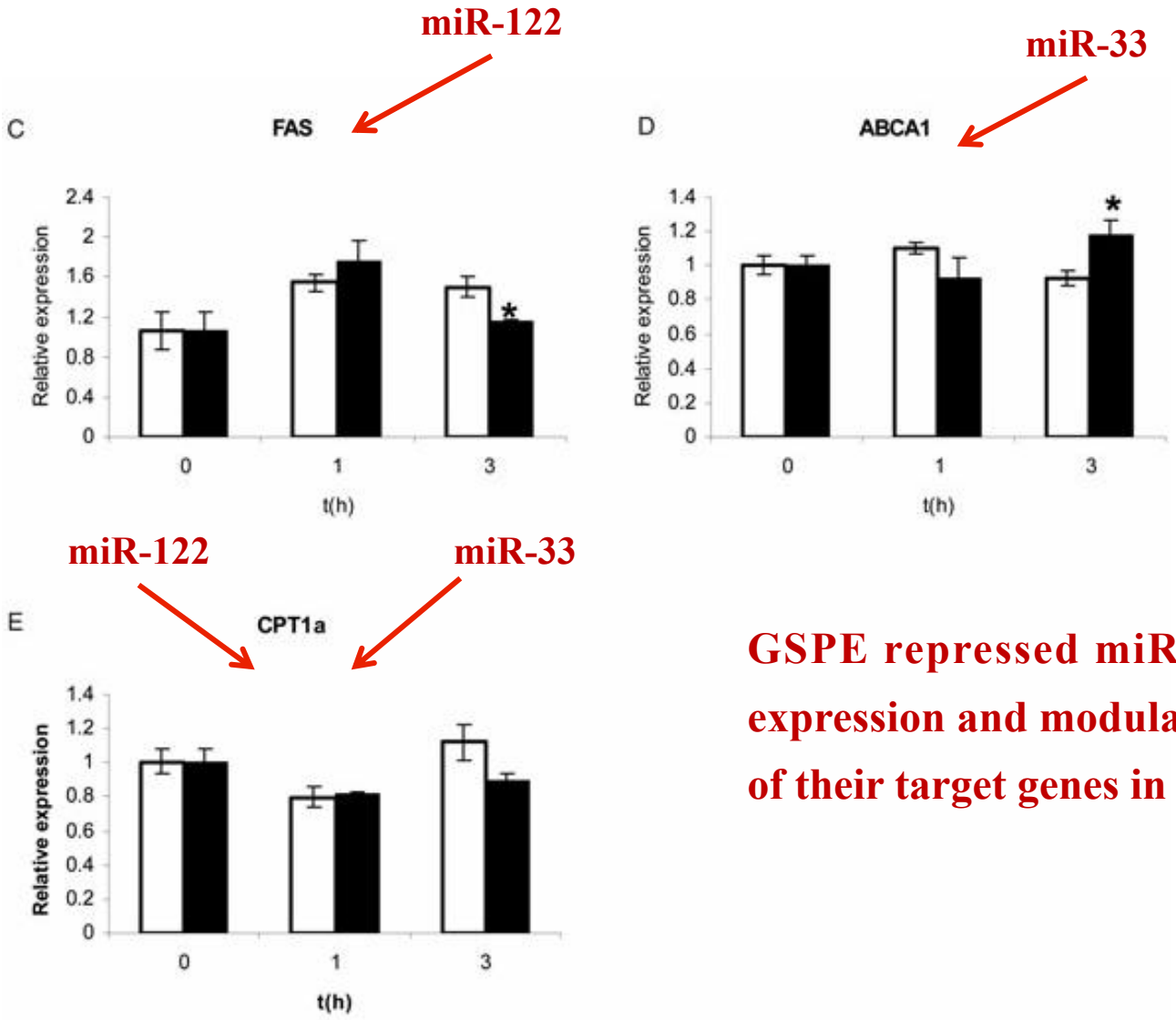


Figure 1. Levels of miR-122 (A),miR-33 (B), and their target mRNAs (C–E) in the livers of rats fed on lard oil with or without proanthocyanidins .

rapid & transient.



GSPE repressed miR-122 and miR-33 expression and modulated the expression of their target genes in the liver in vivo

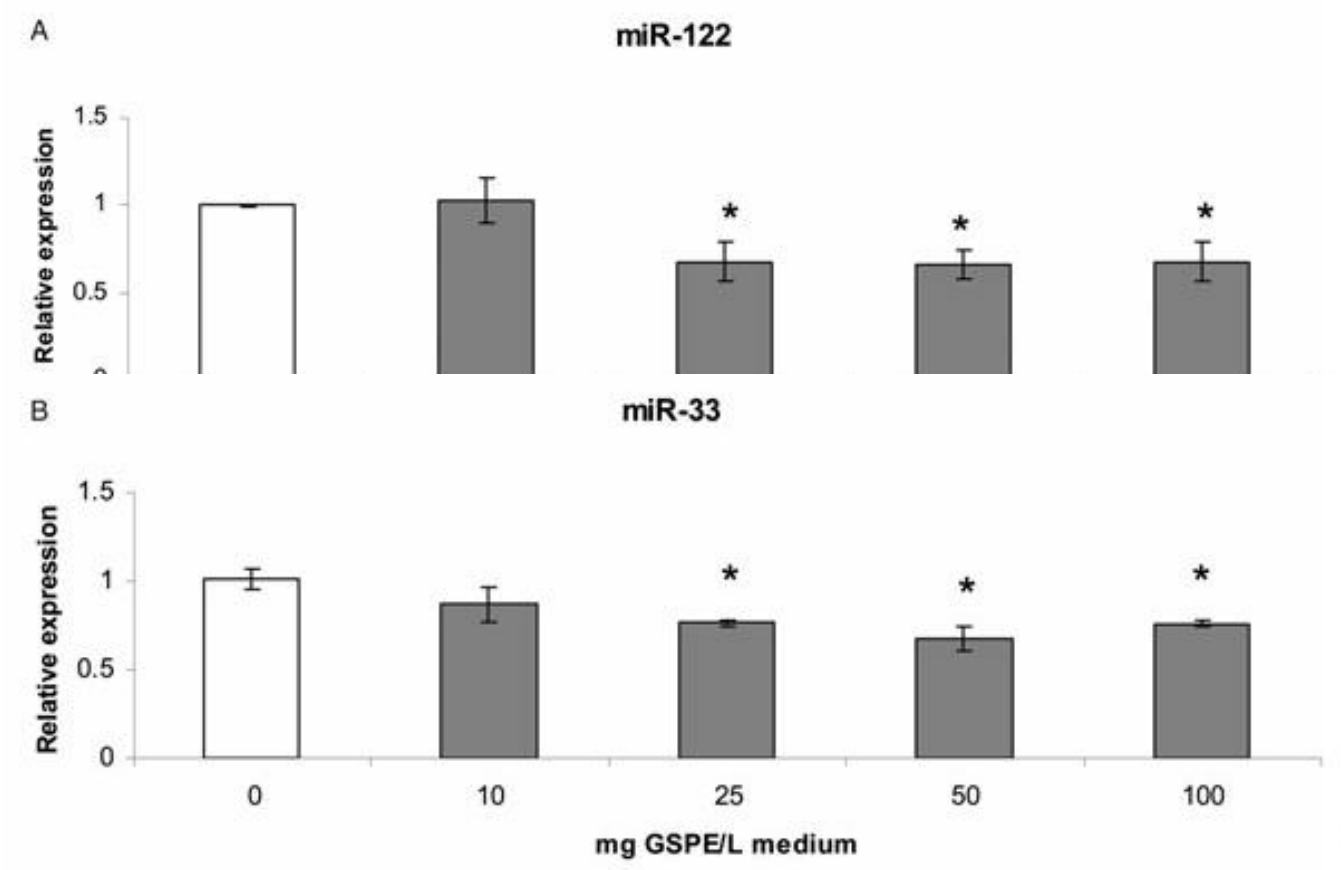


Figure 2. The effect of increasing doses of proanthocyanidin extract on miR-122 (A) and on miR-33 (B) levels in FAO cells.

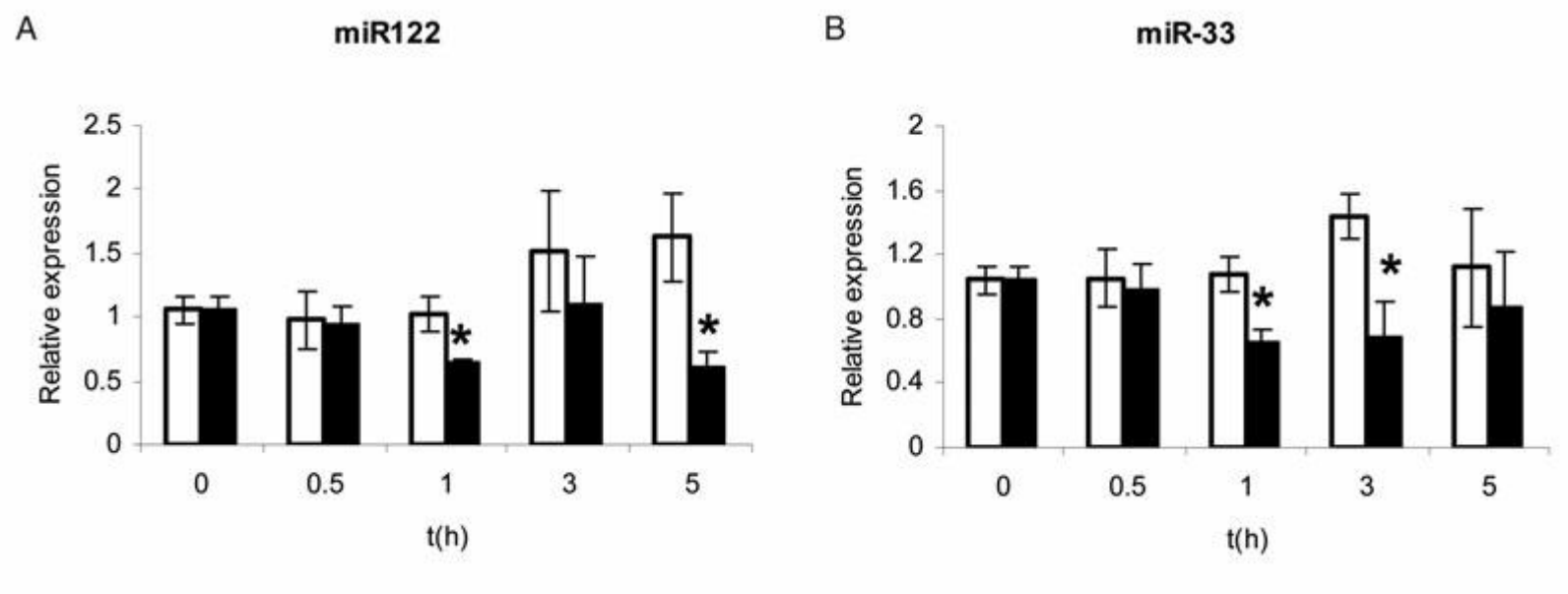


Figure 3. The effects of proanthocyanidin extract on the levels of miR-122 (A), miR-33 (B), and their target mRNAs (C, D) in FAO cells.

constant & not transient

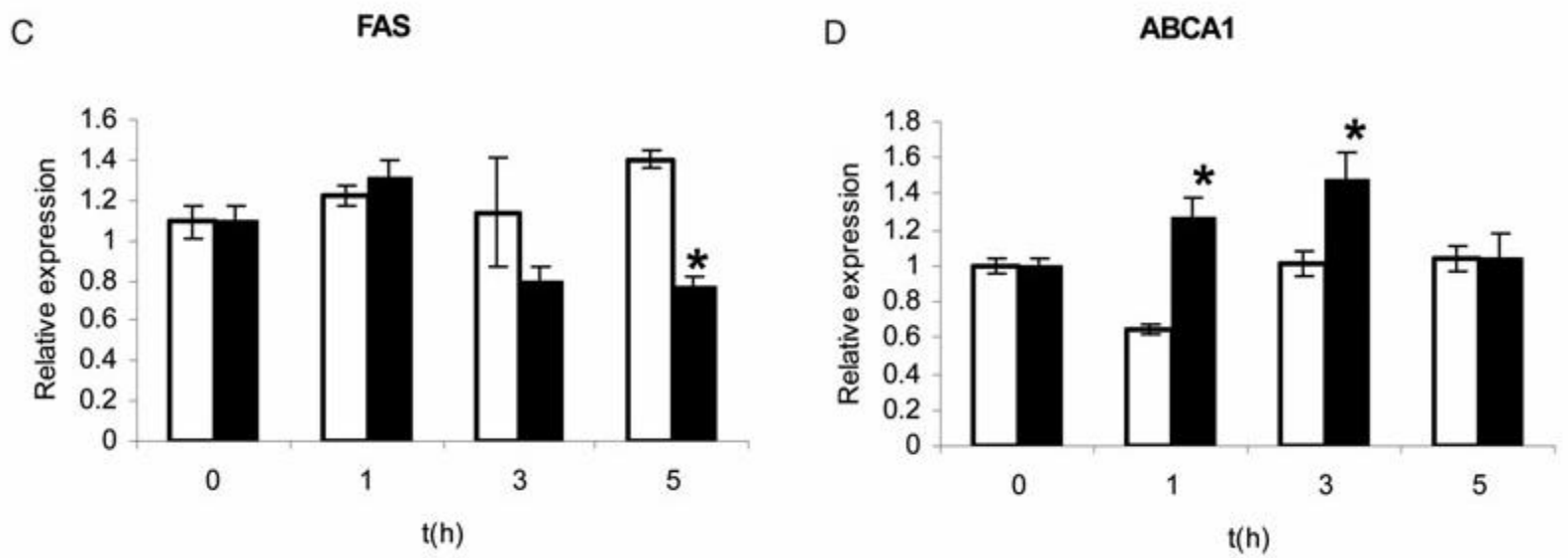


Figure 3. The effects of proanthocyanidin extract on the levels of miR-122 (A), miR-33 (B), and their target mRNAs (C, D) in FAO cells.

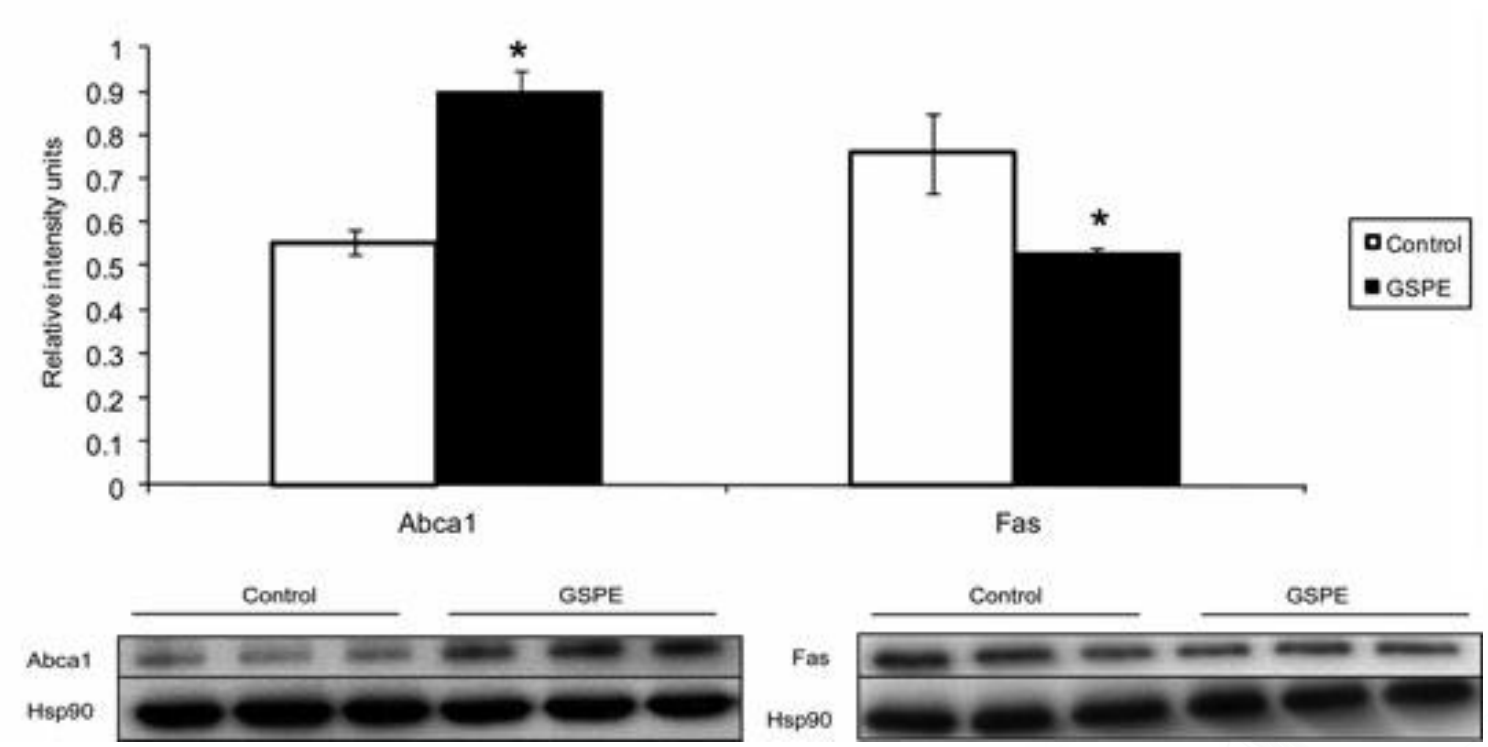


Figure 4. The effects of proanthocyanidin extract on the protein levels of Abca1 and Fas in FAO cells.

Analysis

The molecular mechanism by which proanthocyanidins modulate miRNAs levels is unknown. However, there is evidence that polyphenols can bind to mRNAs and proteins . **Therefore, it is possible that they also bind to miRNAs or to some component involved in miRNA biogenesis, such as DICER or RISC.** More studies will be necessary to identify the mechanism by which GSPE modulate miR-33 and miR-122 levels in liver.

Conclusion

In conclusion, GSPE represses miR-33 and miR-122 in rat hepatic cells, both in vivo and in vitro. The repression of these miRNAs by GSPE is rapid and transient. Moreover, GSPE represses Fas and promotes the expression of Abca1, both of which are target genes of these miRNAs. These data suggest that GSPE increases liver cholesterol efflux to stabilize and promote HDL formation and reduce fatty acid synthesis. Therefore, the repression of miR-122 and miR-33 can be considered a new mechanism of action through which proanthocyanidins exert hypolipidemic effects in the liver.

The End

Thanks for Your Attention