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MINI-REVIEW

The advantages of deep ocean water for the development of functional fermentation food

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Abstract Deep ocean water (DOW) is obtained from 600 m below the sea surface. In recent years, DOW has been applied in the development of fermentation biotechnologies and functional foods. DOW is rich in trace minerals, comprises multiple physiological and health functions, and is able to promote microbe growth; therefore, the application of DOW directly benefits the development of the fermentation industry and functional foods. This study integrated the current health functions and applications of DOW with the latest results from studies related to fermentation biotechnology. Subsequently, the influence of applying DOW in fermented functional food development and the effects in health function improvements were summarized. According to the previous studies, the main reasons for the increased effect of fermented functional foods through the application of DOW are increased generation of functional metabolite contents in the microbes, intrinsic health functions of DOW, and the microbial use of mechanisms of converting the absorbed inorganic ions into highly bioavailable organic ions for the human body. These combined advantages not only enhance the health functions of fermentation products but also provide fermentation products with the intrinsic health functions of DOW.

Keywords Deep ocean water · Functional food · Fermentation · *Monascus* · *Antrodia camphorata*

Introduction

Deep ocean water (DOW) is obtained from 600 m below the sea surface. Seawater in this region is cold and pressurized up to 20 atm, without the interference of any sunlight. In this environment, plankton cannot photosynthesize, water quality is not affected by atmospheric variations, and common microbes cannot survive. Therefore, this region is considered to be near-sterile and rich in inorganic nutrition and various trace minerals. DOW features low temperatures, rich minerals, clean water quality, and balanced amount of trace elements, containing more than 90 types of natural minerals (Othmer and Roels 1973). These minerals are required by the human body but are difficult to obtain from natural food sources alone. The concentrations of these trace elements are higher in DOW than other water sources. Because DOW is undisturbed by sunlight, the water quality is stable, clean, cold, and rich in mineral and nutrient contents (Othmer and Roels 1973). In recent years, numerous studies worldwide have confirmed the physiological functions of DOW (Bak et al. 2012; Hou et al. 2013; Hwang et al. 2009a; Yokota et al. 2010); therefore, its development as a type of functional food is considered feasible. Based on research results related to DOW in the development of preventive health care and applications in fermentation biotechnology, this review summarizes the benefits of applying DOW in the development of fermented functional foods.

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The multihealth benefits of DOW

The multiple health benefits provided by application of rich DOW mineral contents have been proven in numerous studies (Table 1). These health benefits are summarized in the following subsections:

Antiobesity and antidiabetic

Hwang et al. used animal experiments to confirm the role of DOW in antiobesity and antidiabetic effects. In animal experiments, obese mice were fed with DOW on a daily basis for 84 days. The results showed that the final mean body weight

Table 1 The research of the health functions of deep ocean water

Health functions	Functional elements	References
Antiobesity and antidiabetes	Unknown	(Ha et al. 2013; Hwang et al. 2009a)
Prevention of cardiovascular disease	Unknown	(Ha et al. 2014; Yoshioka et al. 2003)
Antihypotensive	Unknown	(Katsuda et al. 2008; Sheu et al. 2013)
Skin protection	selenium	(Bak et al. 2012; Hataguchi et al. 2005; Kimata et al. 2002)
Protection against duodenal ulcers	Mg^{2+}	(Yang et al. 2014)
Osteoporosis prevention	Unknown	(Liu et al. 2013)
Antifatigue effect	Unknown	(Hou et al. 2013)

of DOW-treated mice was decreased by about 7 % compared to control mice. These results indicate that DOW is effective in reducing body weight gain ($p<0.05$). Furthermore, DOW intake also effectively reduced 35.4 % fasting blood glucose levels. The results of oral glucose tolerance test revealed that DOW-fed groups significantly increased the glucose disposal. Additionally, GLUT4 and AMP-activated protein kinase expressions in the musculoskeletal tissues increased because of the intake of DOW (Hwang et al. 2009a). This mechanism provided of DOW primarily consisted of suppressing the mRNA expressions of PPARγ, C/EBPα, fatty acid-binding protein, and adiponectin according to the results of cell test. DOW was found to suppress the relevant gene expressions in adipocyte differentiation, which subsequently suppressed mRNA expressions and cell differentiation (Hwang et al. 2009b). Ha et al. (2013) indicated that DOW promoted PI3-K, AMPK, and mTOR pathway-mediated glucose uptake and increased AMPK phosphorylation of the C2C12 myotubes. Various DOW samples with distinct hardness levels (1000, 2000, and 4000 ppm) were also fed to streptozotocin (STZ)-induced diabetic mice for 4 weeks, which improved AMPK phosphorylation in the muscles. This effect modulated glucose metabolism and subsequently suppressed high blood glucose and improved glucose intolerance (Ha et al. 2013). These results confirmed the antiobesity and antidiabetic effects of DOW.

Prevention of cardiovascular disease

The application of DOW in the development of the hypolipidemic effect has been increasingly studied in Japan. Yoshioka et al. (2003) conducted a 4-week animal experiment on hyperlipidemia rabbits and found that serum total cholesterol and low-density lipoprotein (LDL) cholesterol increased because of high-cholesterol diets. By contrast, this effect in experimental group rabbits fed with DOW was noticeably suppressed. In addition, the effect of drinking DOW on high-density lipoprotein (HDL) cholesterol content and liver function indicators (alanine transaminase and aspartate transaminase) was negligible (Yoshioka et al. 2003). A physician from Kochi Medical School in Japan studied 70–80-year-old subjects to determine the effects of 3-month continuous DOW intake and found that blood T cells from the subjects with prolonged DOW intake exhibited increased immunities. Additionally, circulation rates were promoted because of increased blood flow in the mesenteric artery, which is considered to prevent cerebral thrombosis and myocardial infarction. Moreover, prolonged DOW intake increased iron content in the blood and alleviated anemia in the older subjects. DOW not only demonstrated hypolipidemic effects but also improved the health functions of fatty livers. Another study indicated that DOW suppresses lipogenesis and cholesterol synthesis in the liver, increases β-oxidation-related gene expression, and improves the phosphorylation of IRS-1, LKB1, AMPK, and mTOR in fat and liver tissues (Ha et al. 2014).

Hypotensive effect

In another hyperlipidemic rabbit model, animal experiment proved the hypolipidemic effect of DOW. Additionally, DOW was found to noticeably improve blood pressure (i.e., systolic and diastolic blood pressures), pulse rate, and arterial pressure; Mg^{2+} content in the serum was increased because of DOW intake; however, these improvement mechanisms may not have been caused by the mineral content of the DOW. Although the DOW exhibited improved effects on cardiovascular diseases, relevant studies remain scarce; therefore, in-depth studies of the mechanisms and effects of DOW are required (Katsuda et al. 2008). In another study, the diluted DOW decreased the systolic and diastolic pressures in spontaneous hypertensive rats in an 8-week experiment. DOW also significantly performed hypolipidemic effect (Sheu et al. 2013).

Skin protection

Two groups of Japanese scholars applied DOW in clinical studies on 33 patients diagnosed with allergic dermatitis. These patients were fed DOW daily for 6 months. The results showed that potassium ions in the hairs of the patients decreased, whereas the selenium content in the hairs noticeably increased. Moreover, DOW intake reduced the Hg and Pb toxic heavy metal content. Regarding dermatitis prevention, the conditions of 27 of the 33 patients drastically improved because of the long-term DOW intake. These studies also proved that DOW demonstrated marked effects in treating skin-related diseases (Hataguchi et al. 2005; Kimata et al. 2002).

Protection against duodenal ulcers

Previous studies have indicated that the selenium content in DOW elevates the expression levels of antioxidative substances and antiapoptotic genes and reduces proapoptotic gene expression in rats. Drinking DOW (hardness 600 and 1200) and selenium-containing water can increase both Bcl-2 and thioredoxin reductase 1 expressions. Drinking DOW has also reduce the ulcer area and apoptotic signaling in acetic acid-induced duodenal ulcers, and selenium has been found to be the primary functional element in these effects (Yang et al. 2014).

Osteoporosis prevention

DOW was proven as the therapeutic potential drink on osteoporosis. Drinking DOW strongly enhanced the bone mineral density and decreased serum alkaline phosphatase activity. DOW-treated bone marrow-derived stromal cells (BMSCs) treated by DOW had higher osteogenic differentiation (including BMP2, RUNX2, OPN, and OCN) and enhanced colony-forming abilities. The study demonstrated the regenerative potentials of DOW on osteogenesis prevention (Liu et al. 2013).

Antifatigue effect

A study investigated the antifatigue effect of DOW on the human body. The results showed that DOW intake restored aerobic power within 4 h; muscle power was restored within 24 h. DOW intake can reduce exercise-induced muscle damage markers (creatine kinase and myoglobin) by reducing oxidative stress. These results proved that DOW can alleviate exercise-induced fatigue and muscle damage (Hou et al. 2013).

Enhanced functional effect of fermentation product when fermented using DOW as the culture water

As mentioned above, DOW has demonstrated hypotensive (Sheu et al. 2013), antidiabetic (Ha et al. 2013), antiobesity (Hwang et al. 2009a, b), and hypolipidemic effects (He et al. 2014; Katsuda et al. 2008; Yoshioka et al. 2003). Presently, the functional food and fermentation industries are closely related. DOW is rich in minerals and trace elements. Numerous traditional fermentation industries have applied these minerals and trace elements to increase the efficiency and quality of fermentation productions. DOW can promote microbe growth, elevate fermentation efficiencies, and increase microbe metabolite production (Wang et al. 2013a). Regarding applications in fermentation technologies, mushroom, yeast, koji, and prebiotic yields are increased by adding appropriate amounts of DOW in the culture media. Therefore, through integrating the advantages of the intrinsic health functions of DOW and increased effectiveness in microbe fermentation, DOW applications are expected to benefit the development of multifunctional foods. Furthermore, DOW can be used to increase fermentation efficiency, reduce fermentation costs, and drastically increase product values and effectiveness. In recent years, DOW has been increasingly applied in the production of fermented functional foods, and our previous studies have proven them to increase the health functions of functional microbial fermentation products. As shown in Table 2, DOW has demonstrated hypolipidemic (Lee et al. 2011) and antiobesity effects (Wang et al. 2013b) through the application of RMD and liver cirrhosis prevention effects through the application of *Antrodia camphorata*-fermented products (Wang et al. 2013a). Therefore, these studies were analyzed to assess the feasibility of prospective DOW applications in fermented functional foods.

Enhanced hypolipidemic effect of red mold dioscorea (RMD) when fermented using DOW as the culture water

In 2011, we used DOW and reverse osmosis water (ROW) as water sources in the fermentation of RMD for the experimental and control groups in hypolipidemic assessments, respectively. We also used Syrian golden hamsters with high-cholesterol diets in an animal model for blood lipid assessments to evaluate the influence of RMD, fermented using DOW as a water source, on blood lipids. The results showed that the RMD-fermented with DOW (DOW-RMD) exhibited higher hypolipidemic effects than RMD-fermented with ROW (ROW-RMD) (Table 2). Subsequently, high-performance liquid chromatography was used to analyze the compositions, showing that DOW-RMD exhibited comparatively high monascin production. Monascin and ankaflavin are both *Monascus*-fermented yellow pigments. Therefore, the two pure substances of monascin and ankaflavin in the DOW-RMD were used to evaluate hypolipidemic effects. The results showed that the total cholesterol, triglyceride, and LDL cholesterol contents in the blood obtained from the monascin and ankaflavin pigments in the experimental group were reduced. This indicated that the two pure substances of monascin and ankaflavin are hypolipidemic functional

Table 2 Effect of culture water (deep ocean water and reverse osmosis water) on the functional effects of the fermentation products

Functional effects	Fermentation product	Cultured with DOW	Cultured with ROW	Reference
TC decreased rate (%)	RMD	24.1	14.0	(Lee et al. 2011)
TG decreased rate (%)	RMD	50.5	42.7	(Lee et al. 2011)
HDL-C increased rate (%)	RMD	19.9	7.5	(Lee et al. 2011)
LDL-C decreased rate (%)	RMD	29.7	23.4	(Lee et al. 2011)
Aorta plaque decreased rate (%)	RMD	64.3	37.3	(Lee et al. 2011)
Fat pads decreased rate (%)	RMD	17.4	8.0	(Wang et al. 2013b)
Weight gain decreased rate (%)	RMD	9.5	-3.3	(Wang et al. 2013b)
Lipolysis increased rate (%)	RMD	17.5	11.9	(Wang et al. 2013b)
Lipogenesis decreased rate (%)	RMD	23.9	6.5	(Wang et al. 2013b)
AST decreased rate (%)	AC	15.5	5.1	(Wang et al. 2013a)
ROS decreased rate (%)	AC	26.3	18.2	(Wang et al. 2013a)
TNF-α decreased rate (%)	AC	50.4	20.1	(Wang et al. 2013a)
α-SMA decreased rate (%)	AC	93.2	38.8	(Wang et al. 2013a)

RMD Monascus purpureus-fermented red mold dioscorea, *AC Antrodia camphorata*-fermented product

compounds and can noticeably reduce lipid peroxidation of the serum, aortic lipid accumulation at the heart, and ratios between LDL-C and HDL-C. Moreover, unlike the positive control group that was fed with monacolin K, the group fed with monascin and ankaflavin did not show a substantial reduction in the blood HDL-C content (Lee et al. 2011).

In another study of high-energy diet-induced rat, obesity and hyperlipidemia showed that triglyceride concentrations of the DOW- and ROW-RMD groups were both significantly lower than those of the high-energy diet group ($P<0.05$) and that the triglyceride concentration of the DOW-RMD group was significantly lower than that of the ROW-RMD group ($P<0.05$). Moreover, monascin and ankaflavin both demonstrated significant triglyceride reduction effects ($P<0.05$). The resulting HDL-C and LDL-C concentrations showed that DOW- and ROW-RMDs did not reduce HDL-C but did reduce LDL cholesterol content, thereby preventing cardiovascular diseases (Wang et al. 2013b).

Enhanced antiobesity effect of RMD when fermented using DOW as the culture water

A previous study investigated the body fat-lowering effect of DOW-RMD by using cell tests and animal experiments. DOW-RMDs exhibited superior effects to ROW-RMDs in reducing weight gain, food intake, food utilization rates, body fat, size and area of adipocytes, triglyceride content in blood serum, and total cholesterol increase in the liver (Wang et al. 2013b) (Table 2). Although previous studies have confirmed the body fat reduction effects of red yeast rice, the effects of RMD were not obvious (Chen et al. 2008). Wang et al. (2013b) revealed that ROW-RMD did not significantly reduce weight gain, fat pads, or the size and number of adipocytes, whereas a significant reduction was achieved by the application of DOW-RMD. In vitro tests further showed that this effect can be explained by the superior suppression in the preadipocyte differentiation function and the lipoprotein lipase (LPL) activities of mature adipocytes (Wang et al. 2013b).

DOW is a rich ion source, exhibits physiological functions, and facilitates fermentation. The replacement of DOW by using ion-rich synthesized water (SW) has been a constant research focus. Our previous studies compared SW with DOW. The SW contained Mg^{2+}, Ca^{2+}, Na^{+}, K^{+}, Fe^{2+}, and Zn^{2+}, which are all primary ions in DOW (Wang et al. 2013b). Through synthetic preparation, these ions in the SW were set at identical concentrations as in DOW. Although the SW was unlike DOW, which contains dozens of ion types, the SW resulted in significantly increased monascin and ankaflavin production in the RMD despite an inferior result in increasing ankaflavin content. In addition to the similar regulatory effects in ankaflavin production, the SW and DOW exhibited numerous variations in the adipogenesis regulation functions of adipocytes. The SW suppressed PPARγ and C/EBPα expressions and increased lipolysis but did not exhibit lipolysis functions. DOW did not suppress PPARγ and C/EBPα expressions but increased lipolysis and suppressed heparin-releasable (HR) LPL activities. DOW-RMD and SW-RMD both suppressed PPARγ and C/EBPα expressions and increased HR-LPL activities. However, the similar results may have been caused by differing factors. DOW-RMD contained relatively high monascin and ankaflavin contents and DOW accumulated in the fermentation product. Therefore, the comparatively high monascin and the six accumulated functional ions in SW-RMD contributed to the suppressive effects (Wang et al. 2013b).

Enhanced hepatoprotective effect of *Antrodia camphorata*-fermented product when fermented using DOW as the culture water

The hepatoprotective functions of *A. camphorata* have been previously proposed. *A. camphorata* polysaccharide exhibits antihepatitis B virus activities and is free of cytotoxicity. In particular, a dose of 50 μg/mL of *A. camphorata* B86 strongly suppresses the functions of surface antigens in the hepatitis B virus and demonstrates effects superior to an application of 1000 unit/mL of interferon-α (Lee et al. 2002). Another study also showed improved conditions in mice with CCl_4-induced chronic hepatitis that were fed with 1 g/kg BW of *A. camphorata* fermentation filtrate (Lu et al. 2011). The aforementioned studies showed that the *A. camphorata* fermentation product exhibits hepatitis prevention effects; however, studies on the alleviation of liver fibrosis remain scarce. According to previous studies, *A. camphorata* cultured in ROW (ROW-AC) ameliorates thioamide (TAA)-induced liver fibrosis, as indicated by marked suppression in the TAA-induced methylenedioxyamphetamine (MDA) and reactive oxygen species (ROS) contents and reduction of inducible nitric oxide synthase expression levels. However, only slight improvements were observed in TAA-induced aspartate aminotransferase (AST) activities, superoxide dismutase (SOD) activities, glutathione reductase activities, and TNF-α and α-SMA expression levels. These results also indicate that, despite the hepatoprotective functions of ROW-AC, the improvement effects were less noticeable in cases of severe liver fibrosis. When DOW is used in submerged fermentation processes, *A. camphorata*-fermented products demonstrate strong liver fibrosis prevention effects. DOW-AC demonstrates stronger liver fibrosis prevention effects in TAA-induced hepatomegaly, increased AST activity, and reduced ROS, TNF-α, and α-SMA levels, compared with ROW-AC (Table 2). In histochemical staining and collagen staining results, DOW-AC exhibits superior condition improvements than ROW-AC does (Fig. 1). This shows that the *A. camphorata* cultured in liquid phase, using DOW, exerts increased preventive effects against liver fibrosis (Wang et al. 2013a).

The hypothesis of DOW on enhancing functional effect

Increasing functional metabolite production

In the fermentation of RMD, DOW was proven as the stimulator for the production of monascin and ankaflavin (Lee et al.

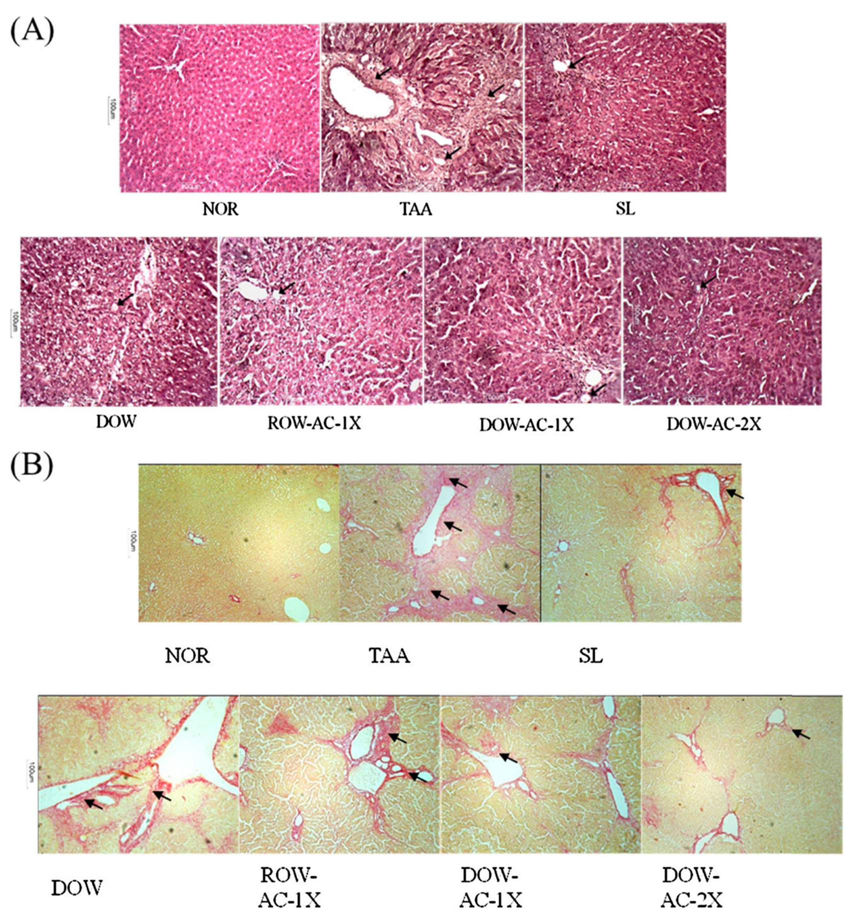

Fig. 1 The effects of *Antrodia camphorata*-fermented product cultured with DOW or ROW on the pathological changes (**a**) and on the collagen accumulation (**b**) in liver tissue of the TAA-induced fibrosis rats. *NOR* normal group. *TAA* TAA-induced fibrosis rats (TAA 100 mg/kg/i.p, three times per week); *SL* TAA-induced liver fibrosis rats fed 100 mg/kg/day of silymarin. *DOW* TAA-induced liver fibrosis rats fed 10-fold concentrated DOW (1.138 mL// kg/day). *ROW-AC-1X* 1-fold dosage of *Antrodia camphorata* product fermented using ROW (63.22 mg/kg/day of mycelium and 1.138 mL/kg/day of 10-fold concentrated filtrate). *DOW-AC-1X* 1-fold dosage of *Antrodia camphorata* product fermented using DOW (63.22 mg/kg/day of mycelium and 1.138 mL/kg/day of 10-fold concentrated filtrate). *DOW-AC-2X* 2-fold dosage of *Antrodia camphorata* product fermented using DOW (126.44 mg/kg/day of mycelium and 2.276 mL/kg/day of 10-fold concentrated filtrate) (Wang et al. 2013a)

2011; Wang et al. 2013b). The two substances of monascin and ankaflavin are functional ingredients in hypolipidemic and antiobesity effects (Lee et al. 2013a, b). Regarding to hypolipidemic ability, monascin and ankaflavin can substantially reduce serum lipid peroxidation, aortic lipid accumulation at the heart, and the ratios between LDL and HDL cholesterol. Moreover, Lee et al. (2011) reported that, unlike a positive control group fed with monacolin K, experimental groups fed with monascin and ankaflavin did not exhibit marked reductions in blood HDL cholesterol content (Lee et al. 2013a). Regarding to antiobesity ability, monascin and ankaflavin suppress the differentiation of 3 T3-L1 preadipocytes in which the expressions of C/EBPα, C/EBPβ, C/EBPδ, and PPARγ are primarily suppressed. Additionally, monascin and ankaflavin were also found to reduce the intracellular accumulation of triglycerides, suppress the lipogenesis of mature adipocytes, and promote the breakdown of intracellular lipid droplets and the subsequent release of glycerin. Additionally, these two ingredients reduced extracellular LPL activities, thereby reducing in intracellular triglyceride synthesis (Jou et al. 2010). In another animal experiment, monascin and ankaflavin were proven to suppress preadipocyte differentiation by suppressing the C/EBPα, C/EBPβ, and PPARγ expressions. By reducing the activity of HR-LPL and increasing lipolysis, monascin and ankaflavin also suppressed the lipogenesis of mature adipocytes (Lee et al. 2013b). As mentioned above, higher hypolipidemic effect and antiobesity effect of DOW-RMD should be contributed by the higher monascin and ankaflavin productions stimulated by DOW.

Furthermore, the improved liver fibrosis prevention effect of DOW-AC may also contribute from the higher functional metabolite production. The synthesis of functional metabolites in *A. camphorata* may be enhanced because of the addition of DOW, based on *A. camphorata* containing higher functional ingredients presenting improved liver fibrosis prevention effects (Table 3). In addition, DOW increased the biomass of *A. camphorata* by 141 %, which further increased the metabolite amounts. Based on metabolite analyses, DOW additions substantially increase polysaccharide, total triterpenoid, total polyphenol, and total flavonoid contents in *A. camphorata*-fermented mycelium or filtrate (Wang et al. 2013a). This proves that DOW addition drastically increases the functional compounds of *A. camphorata* and the antioxidative and anti-inflammatory abilities of these ingredients have been proven in previous studies (Hseu et al. 2005, 2008, 2010; Song and Yen 2002). The aforementioned discussions showed that DOW additions enhance hepatoprotective functions by increasing the antioxidative and anti-inflammatory compounds in *A. camphorata*.

Although DOW is proven as the stimulator for fungi growth and functional metabolite production, however, the DOW may not be the optimal water for fungi fermentation. Various ions in DOW may lead to different effects on the metabolite production. The fungi growth may also be inhibited by too much sodium or other ion concentration. Therefore, the optimal concentration and the ion composition ratio of DOW should be investigated for the fermentation condition of each fungi. Furthermore, the desalting technology and ion ratio modification of DOW are important for the application and development of DOW in the functional food and fermentation biotechnology.

Enhanced functional effect contributed by DOW

DOW contains minerals and trace elements such as Mg^{2+}, Ca^{2+}, and K^{+}. Numerous studies have indicated that high Mg^{2+}/Ca^{2+} ratios facilitate preventive effects against cardiovascular diseases. Cohen et al. (2002) indicated that a daily intake of 8.3 g of Mg^{2+} effectively reduces total cholesterol and triglyceride contents in the blood (Cohen et al. 2002). Shahkhalili et al. (2001) found that a daily supplement of Ca^{2+} (0.9 mg/day) results in reduced blood LDL cholesterol content (Shahkhalili et al. 2001). The fermentation process of

Table 3 Effect of deep ocean water on the modulation of the metabolite production

Fermentation products	Metabolites	Increased rate (%)[a]	Reference
RMD	Monascin	27	(Wang et al. 2013b)
RMD	Ankaflavin	31	(Wang et al. 2013b)
RMD	Citrinin	−4.8	(Wang et al. 2013b)
A. camphorata mycelium	Biomass	141	(Wang et al. 2013a)
A. camphorata mycelium	Triterpenoids	8	(Wang et al. 2013a)
A. camphorata mycelium	Polysaccharides	363	(Wang et al. 2013a)
A. camphorata mycelium	Total polyphenols	−2	(Wang et al. 2013a)
A. camphorata mycelium	Total flavonoids	141	(Wang et al. 2013a)
A. camphorata filtrate	Polysaccharides	70	(Wang et al. 2013a)
A. camphorata filtrate	Total polyphenols	101	(Wang et al. 2013a)
A. camphorata filtrate	Total flavonoids	24	(Wang et al. 2013a)

[a] The increased rate = metabolite production cultured in DOW/ metabolite production cultured in ROW × 100 %

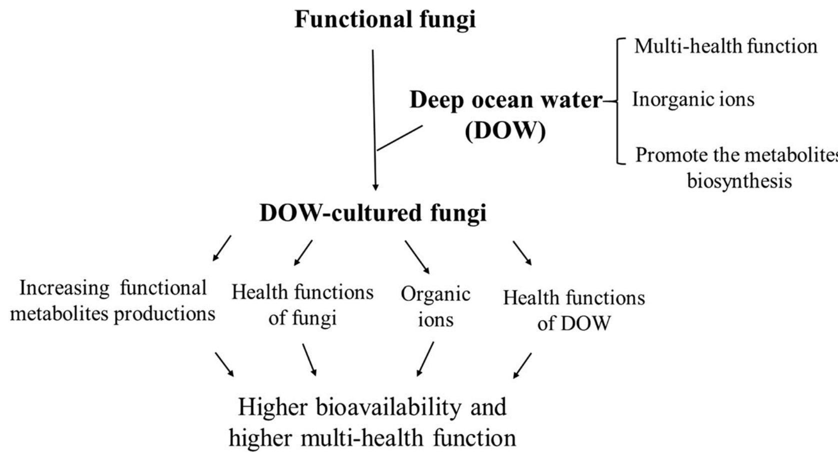

Fig. 2 The advantages and benefits of deep ocean water for the functional effect of functional fungi-fermented products

DOW-RMD requires adding DOW daily. Minerals and trace elements such as Mg^{2+}, Ca^{2+}, and K^{+} gradually accumulating in DOW-RMD facilitate increased hypolipidemic abilities.

The body fat-lowering effect of DOW-RMD has several possible explanations, two of which are summarized in this paper. First, moisture absorption during fermentation retains the functional ingredients of DOW in RMD, thereby exerting a body fat-lowering effect (Lee et al. 2011). A previous study indicated that DOW intake reduces PPAR-γ expression in adipocytes. Hwang et al. (2009b) used DOW in an in vitro cell test to investigate the corresponding influencing mechanism on 3T3-L1 preadipocytes. The results proved that DOW addition effectively suppresses the differentiation of 3T3-L1 preadipocytes, and noticeably reduces the accumulation of lipid droplets and the expression of fat transcription factors (which is also associated with suppressed preadipocyte differentiation) (Hwang et al. 2009b).

Regarding to the enhancement of liver protection by DOW, DOW has intrinsic hepatoprotective functions (Ha et al. 2014; Siscar et al. 2014). In the previous study, the submerged culture consists of DOW as the main water source; therefore, the functional ingredients of DOW were accumulated in the *A. camphorata* mycelium and fermentation filtrates (Wang et al. 2013a). The results also confirmed that DOW exhibited slight preventive effects against liver fibrosis. These included the change in AST activities and reductions in the ROS and malondialdehyde content. Although these effects were not as strong as those of DOW-AC, they were stronger than those of the TAA-induced liver fibrosis group (control). Additionally, DOW substantially reduced TAA-induced TNF-α and α-SMA expression levels and collagen production abilities (Wang et al. 2013a). Previous studies have demonstrated the anti-inflammatory and hepatoprotective effects of DOW (Ha et al. 2014; Siscar et al. 2014). Therefore, the hepatoprotective effects of DOW were accumulated in DOW-AC, which enhanced preventive effects against liver fibrosis (Wang et al. 2013a).

Organic ions converted from the inorganic ions of DOW by microbes

Although DOW is rich in ions and minerals, these ingredients are inorganic ions. Organic compounds such as selenium-methionine are the metabolic products generated through the absorption of biological bodies and the combination with organic compounds (Briens et al. 2013; Du et al. 2008). Previous studies have indicated that inorganic elements are converted into organic forms through utilization by microbes. These organic forms of ions are comparatively easily absorbed by the human body. This technology is the most commonly applied in the fermentation of organic selenium yeast (Se-yeast) (Briens et al. 2013). Numerous studies have used fungi and microbes such as yeast and mushrooms to absorb inorganic Se and convert it into organic Se, which is not only toxic-free but bioavailable to the human body (Du et al. 2008; Stabnikova et al. 2005; Zhao et al. 2004). Therefore, organic Se can be applied to achieve antioxidation and cancer prevention functions (Ahmad et al. 2012; Clausen and Nielsen 1988). DOW comprises multiple trace elements essential to the human body. These inorganinc elements can be converted into organic forms to increase human absorption and intrinsic ionic health functions by using the microbe metabolic process. Previous studies have indicated that DOW-RMD exhibits higher Mg^{2+} concentrations than ROW-RMD does. These ions are absorbed by the *Monascus* and accumulated in fermented products. Although the final product forms of the Mg^{2+} have not been confirmed in current, inferences indicate that *Monascus* may absorb $Mg^{2+}$from DOW, yielding organic forms of Mg^{2+} that may exhibit high bioavailability and enhanced health functions. However, these studies involved in the bioconversion of ion forms in DOW have not been investigated in current, but it should be an important field for raising the bioabsorption and bioavailability of DOW ions in future.

Conclusion

DOW is rich in minerals and trace elements. Two primary factors can be summarized from the aforementioned studies to explain how DOW enhances fermented functional foods (Fig. 2). First, DOW increases functional metabolite production, and increased functional ingredients increase health functions. Second, DOW is rich in ions required in various physiological functions and has been proven to exhibit multiple health functions. The added ions during fermentation processes accumulate in the fermentation products, thereby improving the product health functions. Moreover, inorganic ions are absorbed by microbes during fermentation processes and are subsequently converted to organic compounds in these organisms. Consequently, these biological products become increasingly suitable for absorption and utilization in biological bodies. The synergistic effects of these two factors enhance product health functions. Although DOW enriches the functional ingredients of various fermented functional products, experience regarding DOW application remains insufficient in the fermentation industry and functional food development. The functional food industry is constantly challenged to increase functional effects and research and develop innovative products; the integration of DOW and fermented functional foods provides an innovative industrial development direction that serves these purposes.

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