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Opinion

No Reliable Clinical Results Without a Proper Description of Medicinal Herbs

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Abstract

Complementary and Alternative Medicines (CAM), particularly herbal and natural products (phytotherapy), are gaining popularity, raising concerns about safety and interactions with conventional treatments. With potential benefits, herbs contain bioactive metabolites, necessitating careful consideration of the benefit/risk balance as with other therapeutics. This opinion paper focuses on the critical issue of insufficient herb descriptions in clinical literature, using milk thistle (Silybum marianum) as a case study. The variability in milk thistle extracts, such as Legalon® phytomedicine and other natural food supplements, is substantial, with silymarin being the presumed active extract. However, the literature lacks consistency in reporting extraction methods and metabolite identification. Analyzing clinical studies and case reports related to milk thistle over the past decade reveals significant discrepancies in product descriptions and posology, hindering the establishment of conclusive clinical evidence for its effectiveness.

This opinion article emphasizes the need for a more detailed and standardized approach in describing herbal products in clinical literature. Standardized reporting practices and collaboration between clinicians and phytochemists could be beneficial for bridging the gap between chemistry and clinics in herbal medicine research. The findings underscore the necessity of improved transparency and comprehensive reporting in clinical literature to enhance understanding, interpretation, and reliability in the assessment of herbal medicine efficacy and safety.

Keywords: Complementary And Alternative Medicines (CAM); Phytotherapy; Phytomedicine; Herbal Natural Product; Clinical Study

Abbreviations: CAM: Complementary and Alternative medicines

Introduction

Complementary and Alternative medicines (CAM) are gaining in popularity among patients, with herbs and natural products, known as phytotherapy being especially favored. If these therapies can be beneficial, particularly for minor cures and in some case preventing overuse of conventional drugs, they are often wrongly considered as inherently safe and without any adverse effects. Herbs are known to contain bioactive metabolites. When molecules are bioactive, the balance between benefits and risks must be carefully considered. This implies the potential for interactions with conventional treatments, leading to a loss of clinical effectiveness or the occurrence of toxicities. In certain instances, patients may use herbs as a substitute for conventional treatments, resulting in potentially severe consequences. Understanding the effect of herbs and their interactions with drugs is crucial for clinical practitioners. The ability to identify risks in patients and manage them is necessary to prevent any harmful adverse effects from herbs.

In our opinion, there are several critical points concerning clinical literature on herbal medicine. Since this literature often comes

from clinicians rather than phytochemists/pharmacognosts, the description of the natural health product is often poorly described. It is well known that many environmental factors can influence the qualitative and quantitative composition of secondary metabolites [1, 2]. Moreover, the extraction process significantly contributes to the metabolite bioavailability in patients. For example, thyme essential oil provides access to hydrophobic volatile and fragrant metabolites, while herbal thyme tea provides access to hydrophilic metabolites. Consequently, there is a wide range of extraction methods leading to different compositions. Although the Latin scientific name of herbs is generally described in clinical literature, the extraction method is often omitted. This absence makes it challenging to interpret clinical conclusions accurately.

Herbs in literature

Logically, the main source of information regarding herbs' effect, adverse effects, and interactions is scientific literature. The authors are classically clinicians without proper training in pharmacognosy or sometimes without all information concerning the commercial product used by patients. There are several difficulties for interpreting this phyto-clinical literature. As mentioned above, the mode of extraction is often absent (juice, herbal powder, herbal tea, decoction, maceration... with the extraction solvent and its extractive capacities), and therefore the projection of the type of metabolites is not always accessible. If the literature describes clinical data involving commercial products containing a large number of herbs (as in Asian traditional medicine) or unknown preparation methods (as often in Ayurvedic medicine), it is impossible to rationalize the pharmacological mechanism.

Mentioning only the herb name is not enough. Good redacting practice would require that any herb product mentioned in an article should be easily and undoubtedly identifiable. If for some use cases the scientific name might be sufficient, we believe that going further in herbs description is often required when describing herbs' adverse effects. For example, the herb part used is needed. Examples in clinical literature where herb descriptions are insufficient are numerous; they are particularly present in case reports. To strengthen our proposals, we have chosen to develop the case of milk thistle.

The case of milk thistle

Hepatoprotective phytomedicines or food supplements are often used in addition to a conventional treatment for treating or preventing putative toxic liver damages [3]. Indeed, experiments on cell cultures, animals and humans have shown that natural compounds can prevent or alleviate pathological processes in the liver. For this indication, one herb frequently used is seeds of *Silybum marianum (L.) Gaertn,* [4, 5] commonly known as milk thistle. Botanically speaking, what are commonly called seeds (Figure 1) are the fruit. However, keeping in mind evidence-based medicine standards, final conclusions regarding clinical effects and the effectiveness of this herb are still not established. Despite a large number of trials, clinical evidence remains inconclusive for the effectiveness in treating liver diseases (except in A. *phaloides* intoxications). In our opinion, the main bias explaining the discrepancies in activity is probably due to the different compositions of the extracts tested in these studies and the poor oral bioavailability.

Figure 1: *Fruit of Silybum marianum (L.) Gaertn, named milk thistle seed.*

Indeed, some milk thistle extracts are known to treat liver damage due to toxins, historically due *Amanita phaloides* poisoning (but also many other xenobiotics) and alcohol-induced damage, hepatitis, etc. [6]. In the particular case of milk thistle, there are numerous *in vitro* studies, animal studies, as well as human clinical studies substantiating this use. An intravenous preparations containing silybin (active metabolite – Legalon-sil®) is licensed in Europe, where it is used in emergency rooms for *Amanita phaloides* intoxications. The contents of the bottle correspond to 528.5 mg of C-2',3-dihydrogenic silibinin succinate, di-sodic salt, corresponding to 315 mg of silibinin. After reconstitution with 35 ml of perfusion solution, 1 ml contains 9 mg of silibinin.

In Europe, another oral phytomedicine is available - Legalon®. It is a dry extract of *Silybum marianum* (L.) Gaertn, fruit obtained with a drug extraction (DER) of 36-44:1 equivalent to 140 mg of silymarin (dosed by DNPH or Brady reactive), or equivalent to 108.2 mg silymarin (assayed by LC-UV) calculated on silibinin. It is an ethyl acetate extract with 173.0 - 186.7 mg of dry extract per capsule. There are an innumerable number of other products sold on the market under the status of food supplements with very variable compositions. These products generally contain herb powders for extemporaneous extraction (tea) or direct ingestion, liquid or dry extracts, or purified secondary metabolites isolated from fruits named silymarin.

Milk thistle – chemistry

Silymarin, the supposed "active" extract of this herb, is a standardized extract consisting of approximately 70 to 80 % silymarin flavonolignans generally assayed by LC-UV or LC-MS [7–11]. In numerous bioactive studies, silybin (synonyms: silibinin) is used as standard, as key metabolite. Pharmacologically speaking, it seems to be the most active flavonolignan compound in silymarin.

Silymarin is constituted by:

a. **Flavones:** silybin, silybin, isosilybin and isosilybin A & B (depending on the configuration in C-10 and C-11; $C_{25}H_{22}O_{12}$); silychristin A & B ($C_{25}H_{22}O_{10}$); silymonin ($C_{25}H_{22}O_{9}$); silhermin and neosilhermin A & B ($C_{25}H_{22}O_{9}$)

b. **Flavanols:** silandrin, cisilandrin, isolandrin, isocislandrin A & B, ($C_{25}H_{22}O_{9}$); silydianin and isosilychristin A & B ($C_{25}H_{22}O_{10}$).

c. Flavonoids (taxifolin= dihydro-quercetin $C_{15}H_{12}O_{7}$ and quercetin $C_{15}H_{10}O_{7}$), and the remaining 20-30 % consisting of a chemically undefined fraction (Figure 2) [12].

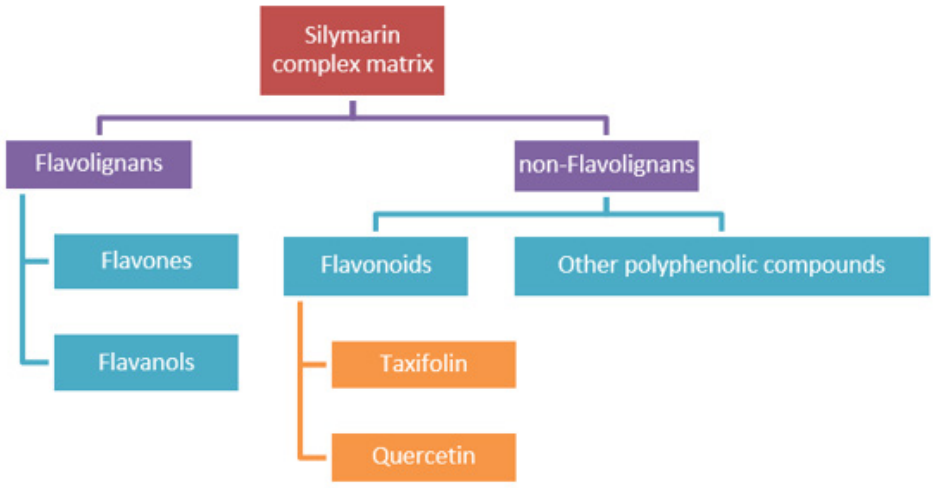

Figure 2: *Overview of silymarin composition from milk thistle seeds.*

The seeds contain about 3 to 6 % of these silymarin extract. As silybin is the main component of silymarin in quantity, the literature mainly focuses on this compound, often ignoring all other components [9]. To our knowledge, 27 compounds have been identified, including all the diastereomers, without considering the "other phenolic compounds" (blue rectangular in Figure 2) [9]. The main component in quantity of silymarin is indeed silybin [13] (in fact a quasi-equimolar mixture of A and B diastereoisomers), and due to this fact biological activity of the whole complex extract is often assigned to both compounds. There are also flavonoids as taxifolin and quercetin (green rectangular in Figure 2). The fruit also contains up to 30 % herbal oil rich in linoleic acid and phytosterols. Supposed active ingredients are not water-soluble, and the preparation of herbal tea and others water solutions seems to make no sense a priori. Low hydro-alcoholic tinctures also appear obsolete. Surprisingly, the EMA in its monograph [14] describes traditional use of different extracts including herbal tea, plant powder or hydroalcoholic tincture with a hepatotropic indication. Effectively, traditional use registration (Article 16a (1) of Directive 2001/83/EC) is described by:

a) No clinical tests and trials on safety and efficacy are required as long as sufficient safety data and plausible efficacy are demonstrated.

b) Involves assessment of mostly bibliographic safety and efficacy data.

c) Must have been used for at least 30 years, including at least 15 years within the EU.

d) Are intended to be used without the supervision of a medical practitioner and are not administered by injection.

To our knowledge, plethoric herbal extracts are sold as herbal food supplements. In the EU, food supplements are regulated as foods and are concentrated sources of nutrients or other substances with a nutritional or physiological effect that are marketed in "dose" form (e.g., pills, tablets, capsules, liquids in measured doses). A wide range of nutrients and other ingredients might be present in

food supplements, including, but not limited to, vitamins, minerals, etc. and various plants and herbal extracts. They are generally standardized organic extracts with silymarin contents of 65 to 80 %, and dosage is determined by the content of silymarin and not by the total volume of extract. Bioactive silymarins are definitely lipophilic and poorly soluble in water, so only about 20–50 % is absorbed from the gastrointestinal tract after ingestion [15, 16]. We believe that one reason why silybin is so popular is because of its very easy method of isolation from silymarin: by precipitation with ethanol from acetone silymarin extract [13].

To support our proposal and our demonstration, we analyzed the phytomedicine Legalon®, an herbal tea that we prepared from fruits, and two herbal food supplements. by LC-MS (procedure will be published soon). The first supplement is a capsule of herbal powder from Arkopharma-France and the second is a hydro-alcoholic (EtOH) extract for NaturActive-France (Figure 3). According to the brand website for the first herbal supplement sold in EU, one capsule contains 390 mg of plant powder with 9 mg of silymarin. It is recommended to take 3 capsules per day at mealtime with a large glass of water, totaling 1170 mg of plant powder or 27 mg of silymarin.

For the second product, the brand website describes that the ethanolic extract contains a "*concentrated extract which is the result of a process aimed at extracting the useful components of the plant in order to increase its virtues tenfold*". The recommended dosage is 2 capsules per day, in the evening at mealtimes, with a large glass of water, i.e., 84 mg of concentrated dry extract of milk thistle fruits. The drug/extract ratio presented is 46 to 70 kg of dried fruits are necessary to obtain 1 kg of concentrated dry extract of organic milk thistle. The exact quantity of silymarin is not specified.

In (Figure 3A), on the left, it is immediately noticeable that the distribution of the different metabolites is completely different according to the commercial products. In phytomedicine (first chromatogram, dry ethyl acetate extract), there are almost as many silychristin A as "silychristin B + silydianin" (peaks around 11 min). On the other hand, in the herbal food supplement (second chromatogram, ethanolic extract), there is three times more silychristin A than "silychristin B + silydianin". In the herbal food supplement based on plant powder (third chromatogram, water at room temperature), the ratio considered is almost respected but the isosylibin A and B are not detectable (peaks around 18 min). Finally, in herbal tea (last chromatogram, water infusion), it is isosilychristin B + silydianin that are absent.

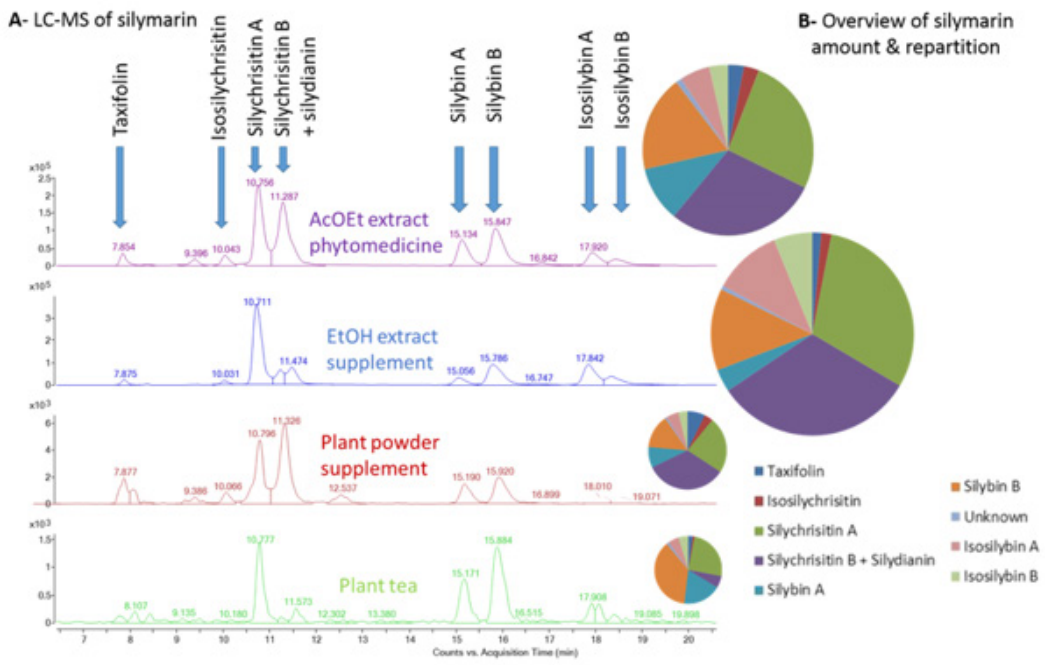

Figure 3: (A) on the left side, 4 chromatograms for the 4 commercial products are presented with the attribution of taxifolin peaks (first peak, $C_{15}H_{12}O_7$) and products identified with the formula $C_{25}H_{22}O_{10}$. Other products with $C_{25}H_{22}O_{12}$, $C_{25}H_{22}O_9$, $C_{25}H_{12}O_7$) are not presented to simplify understanding, (B) on the right side, different pie charts are drawn. The diameter is proportional to the sum of the peak areas. So, it is a representation of the total quantity of metabolites. The parts of different colors represent the distribution of the different metabolites.

But the most striking aspect is in the right part of (Figure 3B). We drew pie charts with the areas under the curve to approach the relative quantities of the different products. The product that provides access to the greatest amount of silymarins is not phytomedicine. Contrary to what one might think intuitively, it is the herbal food supplement obtained with the EtOH extract. The major metabolites in our study are silychristins in 3 of the products and not silybin A & B. In the case of herbal tea, silybin B is the most present.

Milk thistle – clinical data

In these circumstances, it is now clear that clinical evidence of human activity cannot be robust. Especially since the legislation

and controls for food supplements are not as strict as for drugs. We are not certain that we would have obtained the same results by analyzing other batches. The typical oral adult dose of silymarin is 240-800 mg/day in 2 or 3 divided doses [17]. Silymarin, the complex matrix, possesses diverse pharmacological activities, including hepatoprotective. Although clinical trials have shown silymarin is safe at high doses (>1500 mg/day) in humans, the pharmacokinetic (PK) studies of silymarin revealed a poor but rapid absorption (T max = 2-4 h) from oral formulation and its half-life is approximately 6 h. After gastrointestinal absorption, silibinin is rapidly metabolized by phase I and phase II biotransformation in the liver. Around 80% of silibinin is excreted as glucuronide and sulfate conjugates with bile. Around 20–40 % of bile silybin is recovered, whereas the remaining part is excreted via feces. Less than 10 % of orally administrated silybin is excreted in an unchanged form in the urine [18]. At a supratherapeutic concentration (1 µmol/l), silybin had negligible inhibition of the CYP450 enzymes 1A2, 2A6, 2B6, 2C8, 2C9 and 2E1, minor (< 20 %) inhibition of CYP 3A4 and moderate (< 40 %) inhibition of CYP 2C19 and 2D6. Since the therapeutic concentration of silybin is ~ 0.2 µmol/l, authors concluded that silymarin is unlikely to cause hepatic herbal-drug interaction at therapeutic doses [19]. Clinical trials suggest that milk thistle does not affect CYP 1A2, 2C9, 2D6, 2E1, 3A4 or 3A5 [20,21]. In two multiple-dose PK studies, silymarin (160–450 mg every 8 h) did not reduce levels of the CYP 3A4 substrate midazolam, indinavir, rifampin and clarithromycin [22,23]. Although the interaction mechanisms of silymarin via CYP 450 are well studied, there are still many unknowns concerning the pharmacokinetics and impact on CYP. Flavolignans are studied, but the effect of the matrix remains poorly understood, keeping in mind that commercially products have various chemical extracts. In phytotherapy, a *totum* is used, what impact do the "other" metabolites than silymarin have on the absorption of flavolignans? What is the digestion impact? For liver disease, clinical trials on milk thistle studied in an old Cochrane review in 2007 assessed 13 randomized trial (915 patients) have demonstrated safety and efficacy of silymarin (at doses of 1,200-1,500 mg/day) for alcoholic and/or hepatitis B or C diseases [6]. Silymarin have also demonstrated an antidiabetic type-2 effect on patients, reducing levels of glycated hemoglobin, fasting blood glucose, total cholesterol, LDL, triglyceride, SGOT and SCPT compared to placebo in a 4-month treatment in a randomized double-bind controlled trial [24].

To have an overview of how the milk thistle in described in literature, we performed a rapid search on PubMed in November 2023 using the query "milk thistle AND (hepat* OR liver)" and we used the "clinical trial", "randomized clinical trial ", and "case report" filters. We thus limited the search to the 10 previous years (2013-2023). This query resulted in 20 results. Among these articles, we decided to discard 2 studies that focused on horses, 1 was discarded due to being related to a specific case of exposure during professional activity, and we were unable to access 1 other article due to paywall. In the 16 remaining articles, the products were described as follows in (Table 1). As show in these results, only one of the articles presents [36] detailed compositions of the extract used while all others used simpler definition ranging from the name of a supplement (Legalon® in most cases) to the vernacular name 'milk thistle'. Phytomedicine such as Legalon® are likely to always provide the same constituents. Quality must be maintained. One can therefore think that in 8 cases out of 16, the same product is certainly studied. For the other products, it is difficult to draw any conclusion. Posology is consistent through all articles, with daily doses ranging from 140 to 700 mg. The only inconsistencies occur in a case report where doses are lacking, and a metabolic study where a single intake was tested. These differences can be explained by the nature of the article itself.

Table 1: Key information in clinical studies and case reports involving milk thistle products (if found in articles). Information about the product and posology refers to the most precise description we were able to find in the articles.

Study	Product/Posology	Ref
Clinical study on steatohepatitis	Legalon® (Rottapharm|Madaus, Mylan), 420 mg or 700 mg 3 times daily	25
Clinical study on steatohepatitis	Silymarin isolated metabolites, 700 mg 3 times daily	26
Clinical study on steatohepatitis	Legalon® (Laboratorios Takeda S.A., Mexico City (Mexico)), 70 mg once	27
Clinical study - Phase I Dose-Finding in advanced Hepatocellular Carcinoma	Powdered Siliphos (Indena S.p.A, Milan, Italy), 2, 4, 8, or 12 g	28
Case report with treatment of Amanita Mushroom Poisoning	Legalon SIL, "... first given about 96 h after ingestion at the clinical trial loading dose of 5 mg/kg over 1 h and was continued for 3 days at 20 mg/kg/day..."	29
Clinical study on reduction of liver complications of fingolimod in patients with multiple sclerosis	Dried extract of *Silybum marianum* named silymarin produced by Goldaru, Isfahan, Iran, 140 mg once daily	30
Clinical study on hepatoprotection against anti-tuberculosis drug	*S. marianum* capsule, 200 mg, twice a day	31
Case report on steatohepatitis	Legalon® 140mg 3 times daily	32
Case report on reduction of liver enzyme levels in non-alcoholic fatty liver disease	Legalon® 140 2 times daily	33
Case report with abdominal swelling	milk thistle	34
Case report on CYP2C9-mediated warfarin interaction	Milk thistle (200 mg), dandelion (50 mg), wild yam (50 mg), niacinamide (50 mg) and B12 (1000 µg), in the article the galenic form and posology are not mentioned	35

Clinical study on Human Cytochrome P450 Activity	Legalon® 140mg capsules; MADAUS GmbH, Cologne, Germany - Product lot number B0601214) "Each capsule contains 175 mg dried extract of milk thistle achenes, or 140 mg silymarin (Kroll et al., 2007; Javed et al., 2011). Authors have independently analyzed the capsules to confirm the contents of the biologically active constituents (Brinda et al., 2012) contained silybin A (21.2 mg), silybin B (29.5 mg), isosilybin A (11.4 mg), isosilybin B (8.2 mg), silychristin (31.5 mg), silydianin (36.4 mg), and taxifolin (5.9 mg)." 1 capsule 3 times daily	36
Clinical study in obesity	Zn 1 %, Mg 1 % (Purifarma Distribuidora Química e Farmacêutica, São Paulo, Brazil), FOSs 45 % (NutraFlora®, Westchester, Illinois, USA), Se 0.01 %, GOSs 10%, tixosil 5 %, 1.3/1.6-(b-glycosidic bonds) yeast b-glucans (Saccharomyces cerevisiae) 6% (Biorigin, São Paulo, Brazil), and S. marianum (silymarin extract 9%) (SM Empreendimento Farmacê utica LTDA, São Paulo, Brazil). 2 capsules 2 times daily	37
Clinical study in β-thalassemia	Legalon® capsules (Madaus Pharma, Koln, Germany), 140 mg, 3 times daily	38
Clinical study in β-thalassemia	Legalon® (Madaus Pharma), 140 mg, 3 times daily	39
Clinical study in β-thalassemia	Legalon® (Madaus Pharma, Italy), 280 mg daily	40

As a result, (Table 1) describes commercial products that might actually be different from each other in terms of composition while being associated to the same herb. In our opinion, final conclusions regarding clinical effects and effectiveness of this herb, considering the diversity of products sold, cannot be made. We think that the main bias explaining the discrepancies in activity is probably due to the different compositions of the extracts tested in these studies. These discrepant data reinforce the skepticism by the medical profession. If there is a lack of striking effects on the hepatic disease, it can be said with certainty that some extracts of milk thistle have hepatoprotective and hepatoregenerative effects while also increasing the intracellular concentration of glutathione in liver, and consequently the redox state [41].

Conclusion

In the example of milk thistle, we have shown that many commercial products exist with quite variable compositions. We have analyzed in our lab different commercial products of the different classes available in Europe and demonstrated qualitative and quantitative variations. By analysis of the clinical literature of the last 10 years, we observe that the products change that the posology are heterogeneous. This literature is therefore not directly interpretable. The name of the herb is largely insufficient. In our opinion, we outlined here and integrated existing and emerging transdisciplinary knowledge for a better description of natural products from chemistry to clinic.

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Conflict of Interest

No conflict of interest exists.

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