Food Research 9 (1) : 46 - 54 (February 2025)
Journal homepage: https://www.myfoodresearch.com

FOOD RESEARCH

RESEARCH PAPER

Practical concepts to reveal metabolite group types in natural dietary based on ^{1}H and ^{13}C nuclear magnetic resonance spectra: a narrative review

[1,*]Saifudin, A., [1]Maryati, M., [2]Abu Bakar, M.F., [3]Norimoto, H., [4]Tezuka, Y. and [5]Tanaka, K.

[1]*Faculty of Pharmacy, Universitas Muhammadiyah Surakarta, Pabelan, KTS Solo, Jawa Tengah 57102, Indonesia*
[2]*Faculty of Applied Sciences and Technology, Universiti Tun Hussein Onn Malaysia (UTHM), Muar 84600, Malaysia*
[3]*Pura Pharm Japan Corporation, Room 406, 4th FL., JOHO BLDG., 527 Takata, Toyama, 930-0866, Japan*
[4]*Faculty of Pharmaceutical Sciences, Hokuriku University, Ho-3, Kanagawa machi, Kanazawa 920-1181, Japan*
[5]*Department of Pharmacognosy, College of Pharmaceutical Science, Ritsumeikan University, 1-1-1 Nojihigashi, Kusatsu, Shiga 525-8577, Japan*

Article history:
Received: 15 June 2023
Received in revised form: 25 November 2023
Accepted: 12 April 2024
Available Online: 10 January 2025

Keywords:
Chemical shifts,
Metabolite skeleton,
Specific spectra

DOI:
https://doi.org/10.26656/fr.2017.9(1).194

Abstract

Natural products, in particular secondary metabolites, have been progressively becoming one of the most significant topics in food studies. Concise systematical data from *in vitro* to clinical studies have suggested the direct effect of secondary metabolite groups on biochemical interactions related to health status promotion. Hence, revealing the metabolite group from the raw material to the final product is deemed essential to provide quality assurance for an authentic component. Compared to other numerous chemical analysis methods, nuclear magnetic resonance (NMR) spectra data of natural products are seen as able to provide a firm chemical identity of chemical constituents. Since the atomic level of NMR signal data is specific by means of hydrogen and carbon positions, any molecular building blocks are very typical. Moreover, each data point will be a representative fingerprint for each metabolite group. Furthermore, well-prepared NMR spectra data will provide a comprehensive overview of all metabolite groups with unequivocal identity correlated with origin sources. NMR data, therefore, provide undisputable and fixed fingerprinting evidence of metabolite group identity. However, for food investigators and learners of natural metabolites with a relatively inadequate organic chemistry background, implementing this fundamental method becomes a burden and requires tremendous effort. The goal of this review, in turn, was to provide a practical concept to estimate natural compound groups based on ^{1}H and ^{13}C NMR spectral data, in particular, their positions based on the chemical shift signals approach. The methodology of the review commences by providing an overview of the primary biosynthetic frameworks of natural products. This entails elucidating the unique characteristics of each framework and presenting the proton and carbon chemical shifts derived from documented spectra. The determination of natural product group structure can be facilitated by utilizing the chart of putative spectra shown in this article for each specific natural product.

1. Introduction

Over the past two decades, natural products have been progressively becoming one of the most significant topics in food studies. Moreover, in terms of the issue of "back to nature" as the efforts for ailment prevention and health status promotion through dietary or healthy food supplementation, many governments have implemented various programs to encourage their citizens to consume healthy foods mainly derived from terrestrial plant sources or marine products. Those programs have driven state research institutes or private enterprises to search for new potential products and boost commercial products through industrial stages. However, studies on the benefits of natural products for health have been associated with chemical constituents therein, particularly secondary metabolite groups. Numerous reports from basic to clinical research data have suggested the direct effect of secondary metabolite groups on biochemical interactions related to pathological status. Phenolic groups such as tannin and

*Corresponding author.
Email: *azis.saifudin@ums.ac.id*

eISSN: 2550-2166
Published by Rynnye Lyan Resources

quercetin flavonoid, lycopene terpenoid, and stilbene resveratrol, for instance, have been reported to possess strong antioxidant benefits associated with anti-atherosclerosis diseases (Zhang *et al*., 2021). In mixtures, stilbene polyphenol-rich melinjo seed extract has been reported to be capable of stimulating cell longevity via SIRT-1, AMPK, and ATM protein activation (Matsuno *et al*., 2022), inducing brown adipose tissue thermogenesis, and reducing obesity-associated adipose tissue in rats (Yoneshiro *et al*., 2018). A further double-blind placebo-controlled clinical study showed that the administration of these polyphenol groups was associated with an increase in the ratio of high molecular weight to total adiponectin suggested for functional foods to prevent the risk of obesity and metabolic syndromes (Oniki *et al*., 2020). In the flavonoid group, plant extract preparations containing the rich hesperidin flavonoid have been proven clinically able to treat vein circulation problems, in particular, hemorrhoids (Corsale *et al*., 2018). Phenylpropanoids from *Curcuma domestica* and *Alpinia galanga* rhizomes repressed proliferation and induced apoptosis in some endocrine-resistant breast cancer cell lines with a good selectivity index (Da'i *et al*., 2017; Da'i *et al*., 2019; Pradubyat *et al*., 2022). The clinical study of whole extracts of *A. galanga* was proven to improve learning memory (Srivastava *et al*., 2017). As confirmed by *in vivo* studies, phenylpropanoid also plays a role in the neurosystem by preventing dementia in a mouse model (Kojima-Yuasa *et al*., 2016) and transcellular passive diffusion across the blood-brain barrier (BBB) modeling based on a validated parallel artificial membrane permeability assay (PAMPA) (Simon *et al*., 2022). The widely distributed terpenoid groups in plants have been reported to have important pharmacological effects as antineoplastic, anti-inflammatory, and antiviral (Yang *et al*., 2020). In the food industry, natural aromas, either in single or in mixtures, have been playing a fundamental role in enhancing the appetizing flavor of products and their lifetime, usually comprising phenylpropanoids or terpenoid groups (Angane *et al*., 2022). Hence, revealing the metabolite group from the raw material to the final product is very essential to provide quality assurance for an authentic component.

Providing robust identity data for natural components based on chemical groups has been a fundamental consideration. Compared to other numerous chemical analysis methods, nuclear magnetic resonance (NMR) spectra data of natural products are seen as capable of providing a firm chemical identity of chemical constituents. Since the atomic level of NMR signal data is specific by means of hydrogen and carbon positions, any molecular building blocks are very typical. Each data point will also be a representative fingerprint for each metabolite group. Furthermore, well-prepared NMR spectra data will provide a "holistic view" of all metabolite groups with unequivocal identity correlated with origin sources (Kim *et al*., 2011; Valdés *et al*., 2021; Picone *et al*., 2022). Thus, NMR data are affirmative and efficient in providing unambiguous evidence of metabolite group identity. However, for beginner investigators and learners of natural metabolites with a relatively inadequate organic chemistry background, implementing this fundamental method becomes a burden and requires tremendous effort. This review, in turn, aims to provide a practical concept to estimate natural compound groups based on ^{1}H and ^{13}C NMR spectral data, in particular, their positions based on the chemical shift signal approach.

2. Building block concepts of natural metabolites

The original natural products can be tracked on the basis of their building blocks based on biosynthetic pathway origins: acetic acid, mevalonate acid, deoxyglucose phosphate pathway, shikimic acid, and combinations (Dewick, 2002; Bruce and Onyegbule, 2021). The acetic acid pathway results in a C2 building block consisting of fatty acids and polyketide molecules. The former is characterized by a long carbon chain, while the latter possesses an aromatic ring. The second pathway produces the C5 building block with an isoprenoid skeleton. Sometimes, C5 is also called by its terpenoid group from monoterpene to tetraterpene. This group typically has the methyl group branch or its modifications like methine oxide, or exomethylene. Both fatty acid and terpenoid derivatives very rarely have an aromatic ring. Meanwhile, the shikimic acid pathway produced C9, or phenylpropanoid compound, which is characterized by a benzyl ring (C6) with propanoids (C3) as side chains. Propanoid side chains sometimes undergo modification to result in C7. It should be noted here that if polyketide possesses more than one hydroxyl or alkoxyl group, it will be in the meta-position, while phenylpropanoid, in the same condition, will *be* in the ortho-position. For phenylpropanoids, one of these functional groups is at the *para* position towards the propanoid side chain.

Some of these building blocks are those that are capable of conjugating with each other to produce a combined skeleton, such as flavonoid and unhydrolyzed tannins (C2 and C9), panduratins (C2 and C5), cannabinoids (C2 and C9), prenylated coumarins (C5 and C9), or prenylated flavonoids (C2, C9, and C5). These basic building blocks, on the other hand, are also possible to be bound with primary metabolites, especially sugar, to form glycosides, such as ginsenoside saponins (C5 and sugar), hydrolyzed tannin (C7 and

https://doi.org/10.26656/fr.2017.9(1).194

sugar), or flavonoids glycosides. The summary of metabolite building blocks is summarized in Figure 1. Meanwhile, alkaloid groups normally possess building blocks derived from their amino acid origins. Therefore, an alkaloid possessing an aromatic ring or an aliphatic framework will follow those basic rules. Consequently, olefinic functional subgroups are frequently present in aliphatic or cyclic side chains of alkaloids.

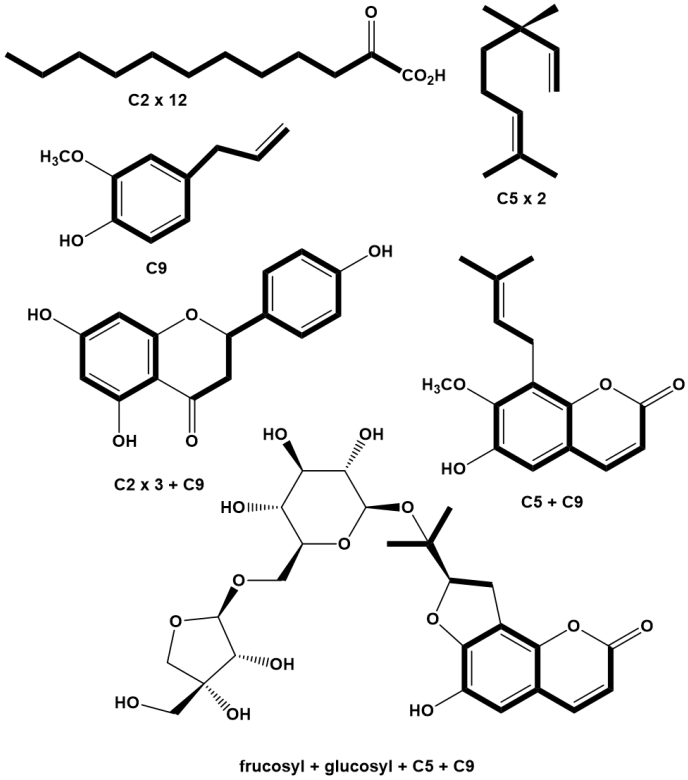

Figure 1. Basic skeletons of natural compounds, as indicated by bold lines depicted according to plausible biosynthesis pathways. Source: Dewick (2002) and Bruce and Onyegbule (2021).

2.1 Acetic acid and terpenoid building block typical signals

The main determinant of hydrogen or carbon signal position is related to the contribution of atomic electronegativity surrounding, in particular, those directly bound to carbon atoms. The more the electronegative functional groups or atoms bound directly to carbon, the bigger the chemical shift in both hydrogen and carbon signal positions (Silverstein *et al*., 2005). Since hydrogen is attached directly to carbon, it will have a similar tendency for chemical shift values; therefore, the hydrogen of primary, secondary, and tertiary alkyls lay in the lower signal position, ranging from d_H 0.5 to 2.5 ppm (Miyake *et al*., 2009; Saifudin *et al*., 2012). While those carbon signal positions, including quaternary carbon, are present at around d_C 8 to 65 ppm, respectively (Ferreira *et al*., 1998). Taken together, these two signals represent alkyls of terpenoid (Li *et al*., 2009) or fatty acid groups (Knothe and Kenar, 2004; Di Pietro *et al*., 2020). Furthermore, at d_H 5-6 ppm, the unsaturated fatty acids are identifiable with vinyl signals (Barison *et al*., 2010). Around d_H 0.5 and 2.5 ppm, respectively, one can observe the carbons of terpenoid groups and typical methyl hydrogen signals. Acetic acid or terpenoid derivatives can sometimes be distinguished based on the presence of methine hydrogen and carbon signals or the presence of singlet methyl signals for terpenoids (Heymann *et al*., 1994; Lee *et al*., 2019). Accordingly, based on the backbone of acetic acid compounds, one can estimate the carbon number as C2 × n or C5 × n. In the case of these metabolite groups possessing common moieties like alkenyl and aldehyde, their hydrogen signals will present at the chemical shifts of d_H 5-6 and 12-13 ppm, respectively. The other common group are carbonyl carbons, whose signals are present at chemical shift values ranging from *dc* 168 to 220 ppm depending on the atom's electronegativity power influencing this carbon. Based on those characteristic chemical shift data values, terpenoids, fatty acids, tocopherols, and other prenylated skeletons, as a consequence, result in signals predominantly in those areas. Another parameter is the signal intensities, which are appreciable with the integration values of 3, 2, and 1 for hydrogen methyl, methylene, and methane, respectively. In the case of unsaturated alkyl chain groups such as those in carotenoids, unsaturated fatty acids, or squalene, their alkenyl signals appear between d_H 5 and 6.5 ppm with the integration values of 1 or 2, dependent upon the attached hydrogen number. For corresponding carbons, their intensities have a similar tendency. Furthermore, quarternary carbon has the lowest intensity. The distribution of those chemical shift signal positions is summarized in Figure 2.

2.2 Polyketide and phenylpropanoid building block typical signals

The typical building block of polyketide and phenylpropanoid groups is the presence of a phenyl ring. Instead of adhering to the electronegativity power effect, hydrogen and carbon in the benzyl ring undergo diamagnetic anisotropy, resulting in high chemical shift values (Silverstein *et al*., 2005). Diamagnetic anisotropy is an effect caused by the pi electrons inducing their magnetic field due to the exposed magnetic field from NMR equipment, which results in deshielded hydrogen and carbon nuclei. Their hydrogen signals are recognizable with the chemical shift signal around d_H 6.5 -8 ppm. Meanwhile, their carbon signals are present at chemical shift values ranging from d_C 110 to 170 ppm. Based on those representative chemical shifts, the aromatic hydrogen of metabolites containing polyketide (Simpson, 1987; Weissman, 2009) or phenylpropanoid skeletons (Liang *et al*., 2006; Fraser and Chapple, 2011) such as coumarins, flavonoids, tannin, xanthones,

https://doi.org/10.26656/fr.2017.9(1).194

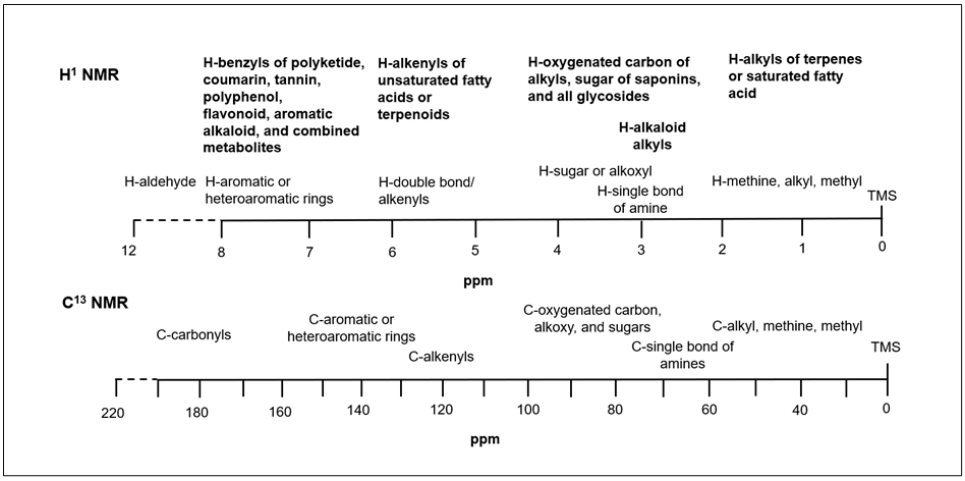

Figure 2. Typical chemical shift position of basic metabolite skeleton group in ^{1}H NMR spectra based on plausible biosynthesis pathways. In the ^{13}C NMR spectrum, each typical metabolite has a similar trend. Usually, tetramethylsilane (TMS) is used as an internal standard.

anthraquinones, cannabinoids, or phenylpropanoid essential oil results in representative benzyl hydrogen and carbon signals at those chemical shift areas. For more specific consideration, the signal intensity of benzylic hydrogen integration is easily noticeable with an integration value of 1 as the shortest signal shape proportionally compared to the others. For its splitting patterns, the signals are basically split by direct adjacent hydrogen in the case of ortho position. Nevertheless, they interact through bounding with farther hydrogen at meta or para positions adhering to the ABK system. These aromatic hydrogen positions can also be determined based on typical coupling constant values of (*J*) 6–10, 1-4, and 0-2 Hz for ortho, meta, and para positions, respectively. Methoxyls frequently become functional groups at the benzyl ring of a polyketide or phenylpropanoid derivative, which can be recognized by their signals as singlets at d_H 3–4 ppm with an estimated integration value of 3.

2.3 Typical signal of metabolites building blocks combination

According to the two above logical features of chemical shift values and signal intensity for both hydrogen and carbon atoms, and furthermore, based on splitting patterns and proportional integration of hydrogen signals, combined metabolite backbones such as flavonoids (C2 and C9) (Li *et al*., 2008), stilbenes (C2 and C7) (Awale *et al*., 2008), hydrolyzable tannins (Yoshida *et al*., 1991) or unhydrolyzable tannin (C2 and C9) (De Bruyne *et al*., 1999) will give dominant signals predominantly at the aromatic area. Accordingly, the carbon signal number of the flavonoid framework in the aromatic area can be estimated as twelve, representing two benzyls. One or two hydrogen signals sometimes will appear et alkyl, alkenyl, or hydroxylated carbon areas, representing hydrogen at carbon numbers 2 or 3 of the flavonoid skeleton. Sometimes, flavonoids are furnished with methoxy groups, which can be recognized by their signals at d_H 3-4 ppm as singlet peaks with an integration value of 3 (Yoon *et al*., 2011). The stilbene group has a number of similar features (Iliya *et al*., 2003; Iliya *et al*., 2022), except for the bridge connecting two benzyls. Among flavonoids, they may also be able to bind covalently to form unhydrolyzed tannin and oligomeric stilbenes. Thus, their derivatives can be logically estimated based on these basic data. While, the other common combined terpenoid is commonly the prenyl group with acetic acid derivatives like cannabinoids (Radwan *et al*., 2011), phenylpropanoid (prenylated coumarins) (Zhao *et al*., 2022), and prenylated flavonoid (Dat *et al*., 2010), their signals can be recognized proportionally at the alkyl area between d_H 0.5 and 2.5 ppm. Meanwhile, in the case of prenyl backbones containing olefinic hydrogens, their signals will appear around d_H 5-6 ppm. The distribution of those chemical shift signal positions is summarized in Figure 2.

2.4 Alkaloid skeleton typical signal

Alkaloids are chemically characterized as plant-derived organic compounds possessing nitrogen atoms. Although originally identified as one natural product, most alkaloid groups have been categorized as strong pharmacologic agents, particularly as psychotic compounds or fungi-contaminating indicators such as pyrrolidine, pyridine, tropane, pyrrolizidine, isoquinoline, indole, piperidine, and quinoline (Zhuge *et al*., 2017; Dusemund *et al*., 2018). Thus, plants containing those alkaloids, in general, have not been recommended as food components. To the best of our knowledge, only *Piper* sp. and *Capsicum* sp. have been recognized as edible plants containing alkaloids. Piper fructus, containing piperine alkaloid (Derosa *et al*., 2016) and *Capsicum* sp., containing capsaicinoids, have been used as spices in many cuisines for centuries (Haq *et al*.,

https://doi.org/10.26656/fr.2017.9(1).194

2021). In the alkaloid structure, the nitrogen atom has a significantly stronger electronegativity effect in the alkaloid group; it will notably contribute to "fingerprinting" toward hydrogen and carbon chemical shifts. On the piperidine, this compound can be recognized based on H2 or H6 signals of piperidine backbone at d_H 2.6-2.8 ppm (Chen *et al*., 2023) and C2 or C6, d_C 58-65 ppm (Symma *et al*., 2021; Choi *et al*., 2022). On the other hand, capsaicinoids are constructed from the combination between the piperidine ring and the phenylpropanoid skeleton (Figure 3). It then has alkenyl and benzyl hydrogen chemical shifts at d_H 4-5 ppm and 6-8 ppm, respectively. Meanwhile, their corresponding carbon signal positions, splitting patterns, and integration have the same order based on chemical environment concepts. Another edible plant containing alkaloids is *Capsicum* sp., also known as the capsaicinoids group, which consists of capsaicin, hydrocapsaicin, norhydrocapsaicin, homohidrocapsaicin, and nonivamide. Their phenyl methanoid group is easily determined based on the information mentioned in the section on phenylpropanoid and terpenoid parts. The specific signal in this group is the H7 signal, which is at d_H 4.25 ppm with a singlet pattern calculated as an integration value of 2 (Valim *et al*., 2019).

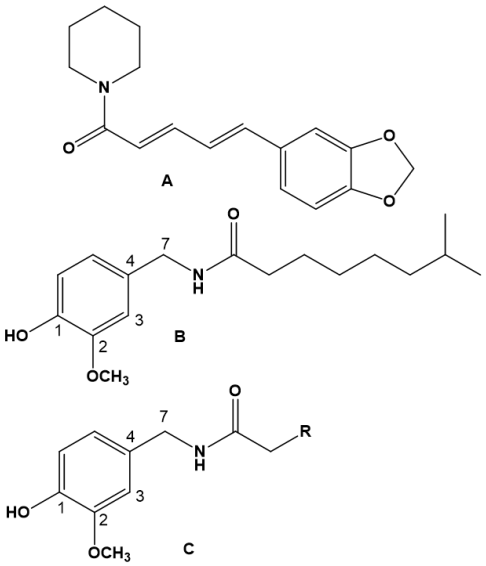

Figure 3. Piperin (A), capsaicin (B), and capsaicinoid derivatives (C) are the only safe alkaloids present in edible plants, *Piper* sp. and *Capsicum* sp. that have been used as spices for centuries.

2.5 Sulphonated skeleton

Sulfur atoms are present in the natural product as functional groups such as thiol, sulphonate, isothiocyanate, and sulfuric acid. The well-known vegetables *Allium sativum*, *A. cepa, and Brassica oleracea* contain relatively little aliin, alicin, and sulforaphane (Figure 4), respectively. The presence of this atom will attract electron clouds toward sulfur atoms. Thus, the chemical shifts of hydrogen and carbon atoms nearby will be more shifted at d_H 2.6-3.5 ppm and their carbon signals will appear around d_C 70-85 ppm (Khiar *et al*., 2009; Recio *et al*., 2018).

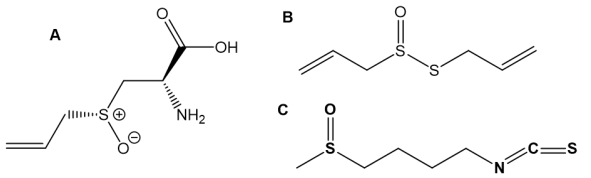

Figure 4. Aliin (A), allicin (B), and sulforaphane. Those sulfonated metabolites reported present limited in edible species such as *Allium* sp. and *Brassica oleracea*.

2.6 Glycoside building blocks typical signals

Glycosides are sugars conjugated to any secondary metabolite group coming from acetic acid, terpenoids, phenylpropanoids, or their combination. The sugar parts usually consist of hexose or pentose molecules, while the non-sugar parts are called aglycons. Their spectra can be recognized by the appearance of hydrogen signals at d_H 3 -4 ppm, which proportionally have the integration value of 1 to those of aglycon parts. Thus, the aglycon part signals either hydrogen or carbon, which can be easily recognized based on the aforementioned understanding concepts. Among them, saponins (a combination of steroids/triterpenes and sugar) will give signals at d_H 2.5-0.5 ppm corresponding to terpenoid skeletons. While flavonoid glycosides' signals are recognized at d_H 7-8 ppm for its aromatic hydrogen parts, glucosynolates give signal d_H 0.5-2.5 depending on their alkyl type. Meanwhile, their ornamenting sugars give around d_H 3-4 ppm. For glucosinolate (Figure 5), which is abundant in the Brassicasseae family, its specific carbon C-7 signal can be observed around d_C 165 ppm (Ibrahim *et al*., 2018). The variability of glucosinolates is determined by the aglycone parts, which can be an aromatic ring, an aliphatic chain, or their combination, so their hydrogen and carbon signals will correspond to each aglycone type. For the fatty acid group, however, there has been no report on its glycosides so far.

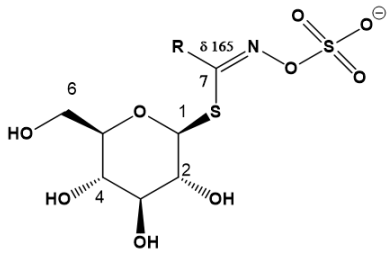

Figure 5. Glucosinolate, a type of glucosides. Its structure with the R site is the chemical shift determinant, its distinguished chemical shift C-7 can be observed around d_C 165 ppm

2.7 Halogenated skeleton

Fluor, chlor, brom, and iodine halogens sometimes

https://doi.org/10.26656/fr.2017.9(1).194

become substituents in a few natural compound groups that have been reported from marine sponges. However, most halogenated compounds have been reported to have an association with environmental toxicity (Ohura *et al*., 2012). The most promising halogenated natural product related to health status is iodinated marine natural products, which have been reported to have antibacterial, antifungal, anti-parasitic, antiviral, antitumor, and anti-inflammatory properties (Gribble, 2015). These halogen substituents in carbon atoms result in the chemical shift value of hydrogen at d_H 4.2-4.8 ppm, while on carbon the effects are complex with fluorine and chlorine generally shifting the carbon resonance to the lower field, but iodine has the opposite influence. All those carbon chemical shifts span on a wide scale. Hence, the previous reports of specific halogen substituents on carbon should be considered for spectral interpretation.

2.8 Solvent contaminant

Samples must be prepared appropriately to avoid any solvent contamination such as well evaporation process. During the extraction steps, organic solvents or a combination of some of them with water are frequently used. Solvent contaminants usually do not have proportional integration with the predominant signals, which are very tall (more than 3 integration values) or very small signals (lower than 0.6). Basically, their signal positions can also be recognized according to the list by Gottlieb *et al*. (1997) and Fulmer *et al*. (2010).

3. Conclusion

The comprehension of the principal features associated with metabolite backbone architectures is facilitated by an examination of the biosynthetic routes of fatty acids, polyketides, terpenoids, and phenylpropanoids. This understanding serves as a foundational step towards comprehending the intricacies of more intricate structures. The identification of hydrogen methyl signals with an integration value of 3, along with the presence of mainly alkyl signals ranging from d_H 0.5 to 2.5 ppm, is indicative of the terpenoid functional group. The subtypes of terpenoids, ranging from mono- to tetraterpenes, can be effectively defined by analyzing their carbon numbers in the ^{13}C NMR spectra. On the other hand, the fatty acid group can be identified by its distinctive carbon carboxylic signal, which typically falls within the range of d_C 170-180 nm. The proton and carbon alkenyl signals of unsaturated fatty acids are observed within the chemical shift range of d_H 5-6 ppm for protons and d_C 120-140 ppm for carbon atoms. The proton and carbon resonance signals associated with polyketides or phenylpropanoid groups are often observed within the chemical shift range of d_H 6.8-8.2 ppm for hydrogen and d_C 125-145 ppm for carbon, respectively. Therefore, a comprehensive comprehension of fundamental spectral data is crucial for the accurate prediction of complex structures found in natural products, including tannins, flavonoids, saponins, stilbenes, and others.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgments

The authors are grateful to Pura Pharm Co. Ltd. for funding the project under the Association for Promoting Sustainable Use of Medicinal Resources (APSUMR) 2021.

References

Angane, M., Swift, S., Huang, K., Butts, C.A. and Quek, S.Y. (2022). Essential oils and their major components: an updated review on antimicrobial activities, mechanism of action and their potential application in the food industry. *Foods*, 11(3), 464. https://doi.org/10.3390/foods11030464

Awale, S., Li, F., Onozuka, H., Esumi, H., Tezuka, Y. and Kadota, S. (2008). Constituents of Brazilian red propolis and their preferential cytotoxic activity against human pancreatic PANC-1 cancer cell line in nutrient-deprived condition. *Bioorganic and Medicinal Chemistry*, 16(1), 181-189. https://doi.org/10.1016/j.bmc.2007.10.004

Barison, A., Pereira da Silva, C.W., Campos, F.R., Simonelli, F., Lenz, C.A. and Ferreira, A.G. (2010). A simple methodology for the determination of fatty acid composition in edible oils through ^{1}H NMR spectroscopy. *Magnetic Resonance in Chemistry*, 48 (8), 642-650. https://doi.org/10.1002/mrc.2629

Bruce, S.O. and Onyegbule, F.A. (2021). Biosynthesis of Natural Products. In Zepka, L.Q., Nascimento, T.C. and Jacob-Lopes, E. (Eds.) Bioactive Compounds-Biosynthesis, Characterization and Applications. InTech Open E-Book. https://doi.org/10.5772/intechopen.97660.

Chen, S.R., Chen, Y.F., Lin, J.J., Ke, T.Y., Lin, Y.S. and Cheng, Y.B. (2023). 2,6-Disubstituted piperidine alkaloids with neuroprotective activity from *Hippobroma longiflora*. *Planta Medica*, 89(3), 308-315. https://doi.org/10.1055/a-1903-2663.

Choi, A., Meijer, A.J., Silvestri, I.P. and Coldham, I. (2022). Kinetic resolution of 2-aryl-4-methylenepiperidines toward enantioenriched functionalizable piperidine fragments. *The Journal of Organic Chemistry*, 87(13), 8819-8823. https://

doi.org/10.1021/acs.joc.2c00862.

Corsale, I., Carrieri, P., Martellucci, J., Piccolomini, A., Verre, L., Rigutini, M. and Panicucci, S. (2018). Flavonoid mixture (diosmin, troxerutin, rutin, hesperidin, quercetin) in the treatment of I–III degree hemorroidal disease: a double-blind multicenter prospective comparative study. *International Journal of Colorectal Disease*, 33(11), 1595-1600. https://doi.org/10.1007/s00384-018-3102-y.

Da'i, M., Suhendi, A., Meiyanto, E., Jenie, U.A. and Kawaichi, M. (2017). Apoptosis induction effect of curcumin and its analogs pentagamavunon-0 and pentagamavunon-1 on cancer cell lines. *Asian Journal of Pharmaceutical and Clinical Research*, 10(3), 373. https://doi.org/10.22159/ajpcr.2017.v10i3.16311

Da'i, M., Meilinasary, K.A., Suhendi, A. and Haryanti, S. (2019). Selectivity index of *Alpinia galanga* extract and 1'-acetoxychavicol acetate on cancer cell lines. *Indonesian Journal of Cancer Chemoprevention*, 10 (2), 95-100. https://doi.org/10.14499/indonesianjcanchemoprev10iss2pp95-100

Dat, N.T., Binh, P.T.X., Van Minh, C., Huong, H.T. and Lee, J.J. (2010). Cytotoxic prenylated flavonoids from *Morus alba*. *Fitoterapia*, 81(8), 1224-1227. https://doi.org/10.1016/j.fitote.2010.08.006

De Bruyne, T., Pieters, L., Dommisse, R., Kolodziej, H., Wray, V., Vanden Berghe, D. and Vlietinck, A. (1999). NMR characterization and biological evaluation of proanthocyanidins: A systematic approach. In Gross, G.G., Hemingway, R.W., Yoshida, T. and Branham, S.J. (Eds.) Plant Polyphenols 2. Basic Life Sciences. Vol. 66. Boston, USA: Springer. https://doi.org/10.1007/978-1-4615-4139-4_10

Derosa, G., Maffioli, P. and Sahebkar, A. (2016). Piperine and its role in chronic diseases. In Gupta, S., Prasad, S. and Aggarwal, B. (Eds.) Anti-inflammatory Nutraceuticals and Chronic Diseases, p. 173-184. Cham, Switzerland: Springer. https://doi.org/10.1007/978-3-319-41334-1_8

Dewick, P.M. (2009). Medicinal Natural Products: a biosynthetic approach. 3rd ed. New York, USA: John Wiley and Sons. https://doi.org/10.1002/9780470742761

Di Pietro, M.E., Mannu, A. and Mele, A. (2020). NMR determination of free fatty acids in vegetable oils. *Processes*, 8(4), 410. https://doi.org/10.3390/pr8040410

Dusemund, B., Nowak, N., Sommerfeld, C., Lindtner, O., Schäfer, B. and Lampen, A. (2018). Risk assessment of pyrrolizidine alkaloids in food of plant and animal origin. *Food and Chemical Toxicology*, 1 (115), 63-72. https://doi.org/10.1016/j.fct.2018.03.005.

Ferreira, M.J.P., Emerenciano, V.D.P., Linia, G.A.R., Romoff, P., Macari, P.D.A.T. and Rodrigues, G.V. (1998). 13C NMR spectroscopy of monoterpenoids. *Progress in Nuclear Magnetic Resonance Spectroscopy*, 33(3-4), 153-206. https://doi.org/10.1016/S0079-6565(98)00022-3

Fraser, C.M. and Chapple, C. (2011). The phenylpropanoid pathway in Arabidopsis. The Arabidopsis Book/American Society of Plant Biologists, 9, e0152. https://doi.org/10.1199/tab.0152

Fulmer, G.R., Miller, A.J., Sherden, N.H., Gottlieb, H.E., Nudelman, A., Stoltz, B.M. and Goldberg, K.I. (2010). NMR chemical shifts of trace impurities: common laboratory solvents, organics, and gases in deuterated solvents relevant to the organometallic chemist. *Organometallics*, 29(9), 2176-2179. https://doi.org/10.1021/om100106e.

Gottlieb, H.E., Kotlyar, V. and Nudelman, A. (1997). NMR chemical shifts of common laboratory solvents as trace impurities. *Journal of Organic Chemistry*, 62(21), 7512-7515. https://doi.org/10.3390/md13074044.

Gribble, G.W. (2015). Biological activity of recently discovered halogenated marine natural products. *Marine Drugs*, 13(7), 4044-4136. https://doi.org/10.3390/md13074044.

Haq, I.U., Imran, M., Nadeem, M., Tufail, T., Gondal, T.A. and Mubarak, M.S. (2021). Piperine: A review of its biological effects. *Phytotherapy Research*, 35 (2), 680-700. https://doi.org/10.1002/ptr.6855.

Heymann, H., Tezuka, Y., Kikuchi, T. and Supriyatna, S. (1994). Constituents of *Sindora sumatrana* Miq. I. Isolation and NMR spectral analysis of sesquiterpenes from the dried pods. *Chemical and Pharmaceutical Bulletin*, 42(1), 138-146. https://doi.org/10.1248/cpb.42.138

Ibrahim, N., Allart-Simon, I., De Nicola, G.R., Iori, R., Renault, J.H., Rollin, P. and Nuzillard, J.M. (2018). Advanced NMR-based structural investigation of glucosinolates and desulfoglucosinolates. *Journal of Natural Products*, 81(2), 323-334. https://doi.org/10.1021/acs.jnatprod.7b00776.

Iliya, I., Tanaka, T., Iinuma, M., Furusawa, M., Ali, Z., Nakaya, K. I., Ali, Nakaya, K., Murata, J. and Darnaedi, D. (2002). Five stilbene glucosides from *Gnetum gnemonoides* and *Gnetum africanum*. *Helvetica Chimica Acta*, 85(8), 2394-2402. https://doi.org/10.1002/1522-2675(200208)85:8%

3C2394::AID-HLCA2394%3E3.0.CO;2-6

Iliya, I., Ali, Z., Tanaka, T., Iinuma, M., Furasawa, M., Nakaya, K.I., Shirata, Y., Murata, J., Darnaedi, D., Mtsuura, N. and Ubukata, M. (2003). Three new trimeric stilbenes from *Gnetum gnemon*. *Chemical and Pharmaceutical Bulletin*, 51(1), 85-88. https://doi.org/10.1248/cpb.51.85

Khiar, N., Werner, S., Mallouk, S., Lieder, F., Alcudia, A. and Fernandez, I. (2009). Enantiopure sulforaphane analogues with various substituents at the sulfinyl sulfur: Asymmetric synthesis and biological activities. *The Journal of Organic Chemistry*, 74(16), 6002-6009. https://doi.org/10.1021/jo9007749.

Kim, H.K., Choi, Y.H. and Verpoorte, R. (2011). NMR-based plant metabolomics: where do we stand, where do we go? *Trends in Biotechnology*, 29(6), 267-275. https://doi.org/10.1016/j.tibtech.2011.02.001

Knothe, G. and Kenar, J. A. (2004). Determination of the fatty acid profile by ^{1}H-NMR spectroscopy. *European Journal of Lipid Science and Technology*, 106(2), 88-96. https://doi.org/10.1002/ejlt.200300880

Kojima-Yuasa, A., Yamamoto, T., Yaku, K., Hirota, S., Takenaka, S., Kawabe, K. and Matsui-Yuasa, I. (2016). 1′-Acetoxychavicol acetate ameliorates age-related spatial memory deterioration by increasing serum ketone body production as a complementary energy source for neuronal cells. *Chemico-Biological Interactions*, 100(257), 101-109. https://doi.org/10.1016/j.cbi.2016.07.031

Lee, T.K., Lee, D., Lee, S.R., Ko, Y.J., Kang, K.S., Chung, S.J. and Kim, K.H. (2019). Sesquiterpenes from *Curcuma zedoaria* rhizomes and their cytotoxicity against human gastric cancer AGS cells. *Bioorganic Chemistry*, 87, 117-122. https://doi.org/10.1016/j.bioorg.2019.03.015

Li, F., Awale, S., Tezuka, Y. and Kadota, S. (2008). Cytotoxic constituents from Brazilian red propolis and their structure–activity relationship. *Bioorganic and Medicinal Chemistry*, 16(10), 5434-5440. https://doi.org/10.1016/j.bmc.2008.04.016

Li, F., Awale, S., Zhang, H., Tezuka, Y., Esumi, H. and Kadota, S. (2009). Chemical constituents of propolis from Myanmar and their preferential cytotoxicity against a human pancreatic cancer cell line. *Journal of Natural Products*, 72(7), 1283-1287. https://doi.org/10.1021/np9002433

Liang, Y.S., Kim, H.K., Lefeber, A.W.M., Erkelens, C., Choi, Y.H. and Verpoorte, R. (2006). Identification of phenylpropanoids in methyl jasmonate treated Brassica rapa leaves using two-dimensional nuclear magnetic resonance spectroscopy. *Journal of Chromatography* A, 1112(1-2), 148-155. https://doi.org/10.1016/j.chroma.2005.11.114

Matsuno, Y., Atsumi, Y., Alauddin, M.D., Rana, M., Fujimori, H., Hyodo, M., Shimizu, A., Ikuta, T., Tani, H., Torigoe, H., Nakatsu, Y., Tsuzuki, T., Komai, M., Shirakawa, H. and Yoshioka, K.I. (2020). Resveratrol and its related polyphenols contribute to the maintenance of genome stability. *Scientific Reports*, 10, 5388. https://doi.org/10.1038/s41598-020-62292-5.

Miyake, K., Tezuka, Y., Awale, S., Li, F. and Kadota, S. (2009). Quassinoids from *Eurycoma longifolia*. *Journal of Natural Products*, 72(12), 2135-2140. https://doi.org/10.1021/np900486f

Ohura, T., Kamiya, Y., Ikemori, F., Imanaka, T. and Ando, M. (2012). Analysis of halogenated polycyclic aromatic hydrocarbons in the air. In Badilla, G.L., Valdez, B. and Schorr, M. (Eds.) Air Quality-New Perspective. InTech Open E-Book. https://doi.org/10.5772/48720

Oniki, K., Kawakami, T., Nakashima, A., Miyata, K., Watanabe, T., Fujikawa, H., Nakashima, R., Nasu, A., Eto, Y., Takahashi, N., Nohara, H., Suico, M.N., Kotani, S., Obata, Y., Sakamoto, Y., Seguchi, Y., Saruwatari, J., Imafuku, T., Watanabe, H., Maruyama, H., Kai, H. and Shuto, T. (2020). Melinjo seed extract increases adiponectin multimerization in physiological and pathological conditions. *Scientific Reports*, 10, 4313. https://doi.org/10.1038/s41598-020-61148-2.

Picone, G., Mengucci, C. and Capozzi, F. (2022). The NMR added value to the green foodomics perspective: Advances by machine learning to the holistic view on food and nutrition. *Magnetic Resonance in Chemistry*, 60(7), 590-596. https://doi.org/10.1002/mrc.5257.

Pradubyat, N., Giannoudis, A., Elmetwali, T., Mahalapbutr, P., Palmieri, C., Mitrpant, C. and Ketchart, W. (2022). 1′-Acetoxychavicol acetate from *Alpinia galanga* represses proliferation and invasion, and induces apoptosis via HER2-signaling in endocrine-resistant breast cancer cells. *Planta Medica*, 88(2), 163-178. https://doi.org/10.1055/a-1307-3997.

Radwan, M.M., ElSohly, M.A., El-Alfy, A.T., Ahmed, S.A., Slade, D., Husni, A.S., Manly, S.P., Wilson, L., Seale, S., Cutler, S.J. and Ross, S.A. (2015). Isolation and pharmacological evaluation of minor cannabinoids from high-potency *Cannabis sativa*. *Journal of Natural Products*, 78(6), 1271-1276. https://doi.org/10.1021/acs.jnatprod.5b00065

Recio, R., Elhalem, E., Benito, J.M., Fernández, I. and

Khiar, N. (2018). NMR study on the stabilization and chiral discrimination of sulforaphane enantiomers and analogues by cyclodextrins. *Carbohydrate Polymers*, 187(9), 118-125. https://doi.org/10.1021/jo9007749.

Saifudin, A., Tanaka, K., Kadota, S. and Tezuka, Y. (2013). Sesquiterpenes from the rhizomes of *Curcuma heyneana*. *Journal of Natural Products*, 76 (2), 223-229. https://doi.org/10.1021/np300694a

Silverstein, R.M., Webster, F.X. and Kiemle, D.J. (Eds.) (2005). Spectrometric identification of organic compounds. 7th ed. USA: Wiley.

Simon, A., Nghiem, K.S., Gampe, N., Garádi, Z., Boldizsár, I., Backlund, A., Darcsi, A., Nedves, A.N. and Riethmüller, E. (2022). Stability Study of *Alpinia galanga* constituents and investigation of their membrane permeability by ChemGPS-NP and the parallel artificial membrane permeability assay. *Pharmaceutics*, 14(9), 1967-1967. https://doi.org/10.3390/pharmaceutics14091967.

Simpson, T.J. (1987). Applications of multinuclear NMR to structural and biosynthetic studies of polyketide microbial metabolites. *Chemical Society Reviews*, 16, 123-160. https://doi.org/10.1039/CS9871600123

Srivastava, S., Mennemeier, M. and Pimple, S. (2017). Effect of *Alpinia galanga* on mental alertness and sustained attention with or without caffeine: a randomized placebo-controlled study. *Journal of the American College of Nutrition*, 36(8), 631-639. https://doi.org/10.1080/07315724.2017.1342576.

Symma, N., Bütergerds, M., Sendker, J., Petereit, F., Hake, A., Düfer, M. and Hensel, A. (2021). Novel piperidine and 3, 4-dihydro-2H-pyrrole alkaloids from *Tilia platyphyllos* and *Tilia cordata* flowers. *Planta Medica*, 87(9), 686-700. https://doi.org/10.1055/a-1340-0099

Valdés, A., Álvarez-Rivera, G., Socas-Rodríguez, B., Herrero, M., Ibáñez, E. and Cifuentes, A. (2021). Foodomics: Analytical opportunities and challenges. *Analytical Chemistry*, 94(1), 366-381. https://doi.org/10.1021/acs.analchem.1c04678

Valim, T.C., Cunha, D.A., Francisco, C.S., Romão, W., Filgueiras, P.R., dos Santos, R.B., Borges, W.S., Conti, R., Lacerda, V. and Neto, A.C. (2019). Quantification of capsaicinoids from chili peppers using ^{1}H NMR without deuterated solvent. *Analytical Methods*, 11(14), 1939-1950. https://doi.org/10.1039/C9AY00292H

Weissman, K.J. (2009). Introduction to polyketide biosynthesis. *Methods in Enzymology*, 459, 3-16. https://doi.org/10.1016/S0076-6879(09)04601-1

Yang, W., Chen, X., Li, Y., Guo, S., Wang, Z. and Yu, X. (2020). Advances in pharmacological activities of terpenoids. *Natural Product Communications*, 15(3), 1-13. https://doi.org/10.1177/1934578X20903555.

Yoneshiro, T., Kaede, R., Nagaya, K., Saito, M., Aoyama, J., Elfeky, M., Yuko Okamatsu-Ogura, Y., Kimura, K. and Terao, A. (2018). Melinjo (*Gnetum gnemon* L.) seed extract induces uncoupling protein 1 expression in brown fat and protects mice against diet-induced obesity, inflammation, and insulin resistance. *Nutrition Research*, 58(9), 17-25. https://doi.org/10.1016/j.nutres.2018.06.012

Yoon, H., Eom, S.L., Hyun, J.Y., Jo, G.H., Hwang, D.S., Lee, S.H., Yong, Y., Park, J.C., Lee, Y.H. and Lim, Y.H. (2011). ^{1}H and ^{13}C NMR data on hydroxy/methoxy flavonoids and the effects of substituents on chemical shifts. *Bulletin of the Korean Chemical Society*, 32(6), 2101-2104. Htttps://*10.5012/bkcs.2011.32.6.2101*

Yoshida, T., Ohwashi, W., Haba, K., Ohbayashi, H., Ishihara, K., Okano, Y., Shingu, T. and Okuda, T. (1991). Tannins and related polyphenols of melastomataceous plants. II. Nobotanins B, C and E, hydrolyzable tannin dimer and trimers from *Tibouchina semidecandra* COGN. *Chemical and Pharmaceutical Bulletin*, 39(9), 2264-2270. https://doi.org/10.1248/cpb.39.2264.

Zhang, S., Li, L., Chen, W., Xu, S., Feng, X. and Zhang, L. (2021). Natural products: The role and mechanism in low-density lipoprotein oxidation and atherosclerosis. *Phytotherapy Research*, 35(6), 2945-2967. https://doi.org/10.1002/ptr.7002.

Zhao, C.X., Gao, H., Yu, M., Zhao, J.P., He, B. X., Wu, J.P., Zhang, H.X., Zhang, T. and Zou, Z.M. (2022). ^{1}H-NMR-guided isolation of enantiomeric coumarin-monoterpenes with anti-inflammatory activity from *Gerbera piloselloides*. *Phytochemistry,* 203, 113346. https://doi.org/10.1016/j.phytochem.2022.113346

Zhuge, Y., Liu, Y., Xie, W., Zou, X., Xu, J., Wang, J. and Chinese Society of Gastroenterology Committee of Hepatobiliary Disease. (2019). Expert consensus on the clinical management of pyrrolizidine alkaloid-induced hepatic sinusoidal obstruction syndrome. *Journal of Gastroenterology and Hepatology*, 34(4), 634-642. https://doi.org/10.1111/jgh.14612

