INSTITUT DE FRANCE
Académie des sciences

Comptes Rendus

Chimie

Wern Huay Mah, Nadzran Hafiy Ahmad Nazuan, Wei Sheung Yeap,
Farah Hasyeena Fakharudin, Ibrahima Faye and Cecilia Devi Wilfred

Deciding which is the best ^{1}H NMR predictor for organic compounds using statistical tools

Volume 25 (2022), p. 83-95

https://doi.org/10.5802/crchim.156

Les Comptes Rendus. Chimie *sont membres du*
Centre Mersenne pour l'édition scientifique ouverte
www.centre-mersenne.org
e-ISSN : 1878-1543

Comptes Rendus
Chimie
2022, 25, p. 83-95
https://doi.org/10.5802/crchim.156

Full paper / *Article*

Deciding which is the best ^{1}H NMR predictor for organic compounds using statistical tools

Wern Huay Mah [a], **Nadzran Hafiy Ahmad Nazuan** [a], **Wei Sheung Yeap** [a],
Farah Hasyeena Fakharudin [a], **Ibrahima Faye** [a] **and Cecilia Devi Wilfred** *, [a]

[a] Fundamental and Applied Sciences, Universiti Teknologi PETRONAS, 32610 Bandar
Sri Iskandar, Perak, Malaysia

E-mails: wern_19001600@utp.edu.my (W. H. Mah), nadzran_18002615@utp.edu.my
(N. H. Ahmad Nazuan), wei_19000827@utp.edu.my (W. S. Yeap),
farah_19001641@utp.edu.my (F. H. Fakharudin), ibrahima_faye@utp.edu.my (I. Faye),
cecili@utp.edu.my (C. D. Wilfred)

Abstract. ^{1}H NMR chemical shifts for 30 organic compounds (396 data points) were predicted using four NMR predictor software and compared with the experimental data from SDBS. The NMR predictors involved were MestReNova, ChemDraw, NMRShiftDB and ACD Workbook Suite. Root mean square deviation (RMSD) and mean absolute percentage error (MAPE) were calculated from the data obtained. One-way analysis of variance (ANOVA), Tukey's honestly significant difference (HSD) and t-test were carried out to analyse the statistical significance of the differences between the predictors. The results from the statistical analysis were used to predict chemical shifts of three organic compounds.

Keywords. NMR predictors, Root mean square deviation, Mean absolute percentage error, One-way ANOVA analysis, Tukey's honestly significant difference, t-test.

Manuscript received 5th August 2021, revised 13th December 2021 and 1st February 2022, accepted 1st February 2022.

1. Introduction

Molecular structure elucidation is the determination of chemical structures which can be achieved by using NMR spectroscopy techniques. The molecular environment around a nucleus determines its chemical shift, and its interpretation is important to elucidate the structure of organic molecules. Accurate predictions of NMR chemical shifts with respect to experimental values are highly valuable for structural elucidations. NMR predictions' software primarily rely on coding and computational methods in producing NMR predictions, different programs can use different methods in their internal calculations for predictions. The focus of this study is to evaluate the accuracy of ^{1}H NMR predictions' software using statistical tools namely on MestReNova, ACD Workbook Suite, ChemDraw and NMRShiftDB. MestReNova and ACD Workbook suite were selected based on the similarity of what they both possess i.e., having the conventional hierarchically ordered spherical description of environment (HOSE) as one of their predictors but the latter having neural network (NN) as an additional tool that can interpolate

---

* Corresponding author.

within its trained database. These predictor's ability was compared with ChemDraw, a predictor that only uses a single prediction method: linear additivity rules. Another predictor's ability was compared, i.e. NMRShiftDB that only uses NNs in predicting NMR shifts.

MestReNova prediction is based on two predictors: MestreLab Predictor and Modgraph Predictor [1]. When an NMR prediction is made, each of these predictors will predict chemical shifts with a certain confidence interval based on different methods, for example with HOSE, along with substituent chemical shifts approach for Modgraph predictor, and CHARGE for MestreLab Predictor. These predicted chemical shifts will be combined using a Bayesian method to produce the final chemical shift, which has higher reliability and accuracy than the individual chemical shifts. The concept utilized is known as ensemble learning, which allows multiple machine-learning algorithms to be combined. The concept of ensemble learning is advantageous as the deficiency of one prediction method can be compensated by another prediction method, improving the overall prediction accuracy while reducing the number of outliers. For example, HOSE prediction method, which relies heavily on the size and quality of database, is highly inaccurate when predicting novel compounds, as very few database compounds possess a structure similar to the novel compound [2]. This deficiency is compensated by the substituent chemical shifts approach, which predicts by identifying substructures and substituents in a molecule and assigning base and increment values respectively based on their identity. Utilizing ensemble learning method, a database containing approximately 1000,000 shift values and 3000 available parameters, MestReNova can produce accurate ^{1}H NMR chemical shifts by combining the strengths of each prediction method used while minimizing their respective weaknesses. ACD Workbook Suite, on the other hand utilizes multiple prediction methods which are additivity rules, HOSE, and NN [3]. HOSE can be compromised when there is error present in database values, as these errors will be reproduced in the final predicted chemical shifts [4]. NN is well-known for its ability to generalize data, it can interpolate within its trained database, consequently less dependent on database values, and thus, reducing the possibility of reproducing errors within database in the final predicted value [4,5]. To ensure robustness, generality, and accuracy in NN, various types of descriptors, namely topological, physicochemical, and geometric descriptors (to account for stereochemistry and 3D effects) were used to better match the properties of H atoms to be predicted to those available in the database [6]. A database of approximately 1400,000 experimental ^{1}H chemical shifts ensure robustness and accuracy of ACD Workbook Suite [7]. ChemDraw's predictor, ChemNMR, relies on a single prediction method: linear additivity rules. Linear additivity rules are similar in concept to the substituent chemical shifts approach used in MestReNova and the additivity rules used in ACD Workbook Suite. However, it was found that linear additivity rules are inefficient in chemical shift prediction of molecules that exhibit non-linearity. Even though correction methods are implemented to improve its prediction power, it is still lacking when resonance effects are present in the molecule. Even so, ChemDraw is still reliable because it considers stereoisomerism and is backed by a large database of approximately 700 base values and 2000 increment values based on 4000 parameters [8]. NMRShiftDB utilizes deep NN in predicting NMR shifts, which achieved better precision than the HOSE code approach. When a molecule is predicted, its molecular weight, atom, and proton count will be compared to database molecules. If there are ten or more chemical shifts available, the smallest and largest chemical shift will be taken as confidence limit of the 6-sphere HOSE prediction. However, if values obtained are insufficient for 6-sphere HOSE or confidence limit estimation, the number of spheres used in HOSE will be reduced, leading to significantly decreased HOSE prediction accuracy as it relies heavily on number of spheres [9].

The diversity and variation of prediction methods among the available NMR predictor software in use are quite high and warrant further examination on the reliability and accuracy of these methods in predicting NMR chemical shifts, as well as comparisons and discussions on which method or program produces predictions that most closely matches real-world experimental data. Thirty organic compounds with 396 data points were used. Statistical comparison methods applied were mean absolute percentage error (MAPE), root mean square deviation (RMSD), one-way analysis of variance (ANOVA), Tukey's honestly significant difference (HSD) and t-test. From the data analysis, the best NMR predictor software was determined. The predictor software was then used to predict ^{1}H NMR of recent compounds found in literature.

2. Methods and analysis

The ^{1}H NMR chemical shifts were obtained by drawing and inserting the molecules into the NMR predictor software. The 30 organic compounds are shown in the supporting documents. The predictor's accuracy was evaluated in aliphatic, aromatic, cyclic and polycylic molecules. For aliphatic molecules, additional functional groups in the molecules add to the complexity in the prediction e.g. ester (7), ester and dicarbonyl (9), diols and ether (10) and ester, diols, alkene (11, 12 and 13). Molecules 1, 2, 4 and 13 were selected due to their cyclic conformations. It would be interesting to know whether the selected predictors are able to predict the chemical shifts of the hydrogens on the ring in these selected conformations. Molecules 1, 2 and 4 have ester functionality in the ring or near the ring, which acts as an electron withdrawing group. The ring in molecule 13 has additional complexity as it is linked to a nitrogen atom adjacent to a sulfone. Molecules 22 and 24 have cyclic moieties but whether it is in chair or boat conformation is unknown. It would be interesting to evaluate the predictor's capability for these kinds of molecules. Again, we are curious to know the accuracy of the predictor when the cyclic compound is bonded to ester group in 22 and aromatics functionality in molecule 24. We had also selected a range of other aromatic compounds as seen in molecules 5, 6, 8, 14–21, 25, 26, 29 and 30. Various other functional groups are linked to the aromatic groups from carbonyl and thiol (5), ketone and alcohol (6), ethers and alcohol (8), sulfone (14), phosphate (15), phosphate where one oxygen atom is replaced by sulfur (16), fluorine (17–19), amide (20 and 21), thiol, carboxylic acid and amide (25), polycyclic (26) and heteropolycyclic with imine (29 and 30). The predictor's capability was further evaluated, in the case of fused aromatic rings. The complexity was further heightened with presence of double bonds (3), polycyclic and dicarbonyl (27) and heteropolycyclic and esters (28).

The details of each software and its link are provided in the supporting document. The variation of the experimental proton shifts with predicted values for a molecule (Compound 1) is shown in the graphs in the supporting documents. The comparison of the calculated and experimental ^{1}H NMR chemical shift values for all the compounds are provided in the SI.

MAPE was calculated by using the equations in (1) and (2), while RMSD was calculated by using formula (3).

$$\text{Percentage error (\%)} = \frac{|\text{Predicted value (ppm)} - \text{experimental value (ppm)}|}{\text{Experimental value (ppm)}} \times 100\% \tag{1}$$

$$\text{MAPE (\%)} = \frac{\Sigma\ \text{Percentage error (\%)}}{N} \tag{2}$$

$$\text{RMSD} = \sqrt{\frac{\sum_{i=1}^{N}(x_i - \hat{x}_i)^2}{N}}, \tag{3}$$

where x_i = experimental ^{1}H chemical shift values, $\hat{x}_i$ = predicted ^{1}H chemical shift values, N = total number of values.

MAPE was chosen as a parameter because it normalizes the absolute error between predicted and experimental values over a range of 0% to 100%. This allows comparison between datasets of different scales, as the absolute error of each predicted chemical shift is normalized by the experimental value, and an average is obtained [10]. RMSD, on the other hand, approximates the spread of predicted data from experimental data without restricting its values to any range.

2.1. *Tests of statistical significance between the software*

Three methods were selected to test for the significant difference between collected data, which are one-way ANOVA with a post hoc test, Tukey's HSD, and t-test. The four sets of significance tests are shown in Table 1.

Both t-test and ANOVA determine the significant difference between populations by observing the mean and variance. T-test analyses two populations, whereas ANOVA is more suited for analysis of three or more populations. Thus, ANOVA was used for the first two datasets to compare the four NMR predictor programs. t-test was not repeatedly carried out as it can contribute to significant Type I error (false positive) which causes a true null hypothesis to be rejected. As ANOVA simultaneously compares all four NMR predictors, the rate of Type I error is

Table 1. Datasets and the corresponding significance tests used

No	Dataset	Significance test
1	MAPE of 30 organic molecules from SDBS	ANOVA + Tukey's HSD
2	RMSD of 30 organic molecules from SDBS	ANOVA + Tukey's HSD
3	MAPE of 3 organic compounds from literature	t-test
4	RMSD of 3 organic compounds from literature	t-test

maintained at the significance level of the hypothesis test, $\alpha = 0.05$. One-way ANOVA was used because the analysis had one independent variable: the type of NMR prediction program. Tukey's HSD (post hoc analysis) was conducted alongside ANOVA to determine which specific NMR predictor was statistically different. Similar to t-test, Tukey's HSD utilizes pairwise comparisons as well, however its data is presented in adjusted p values and simultaneous confidence intervals. These representations maintain the rate of occurrence of Type I errors by limiting the family-wise error rate to the significance level used, $\alpha = 0.05$ [11].

T-test was used in the third and fourth dataset where two NMR prediction software were compared, namely MestReNova and ACD Workbook Suite. The interpretation of data was based on two-tailed prediction, as the deviation of predicted chemical shift from experimental chemical shift is not unidirectional.

2.2. *ANOVA and Tukey's HSD*

In performing the following ANOVA and Tukey's HSD, the null hypothesis (H_0) was that all predictor programs are equal in performance. The alternative hypothesis (H_1) was that at least one of the predictor programs is significantly different in performance than the others.

The process of analysis is as follows: ANOVA (omnibus test) was separately performed on the datasets of MAPE and RMSD obtained from observing the deviations between predicted and experimental values. The MAPE and RMSD data of 30 molecules was analysed. ANOVA produces the F value (F), F-critical value (F_{crit}), and P value (P). If both conditions of $F > F_{\text{crit}}$ and $P < 0.05$ are achieved, it indicates that at least one of the programs is significantly different in performance from the others, affirming H_1 and rejecting H_0.

To verify the conclusion from ANOVA and to identify which specific predictor software is significantly different, Tukey's HSD (post hoc analysis) was performed by calculating and comparing the Q-stat value (Q_{stat}) and Q-critical value (Q_{crit}) for each pair combination of predictor programs (pairwise comparisons). For pair combinations where $Q_{\text{stat}} > Q_{\text{crit}}$, it suggests that one program is significantly different from the other in the pair. The performance of which predictor is better or worse can be determined based on their average MAPE or RMSD. Additionally, simultaneous confidence interval (CI) of each pair combination of predictor programs was calculated and a graph was plotted. The difference in performance between the programs is significant if CI does not encompass zero in the graph.

3. Statistical analysis on the NMR prediction

3.1. *RMSD*

The chemical structure of the 30 molecules is provided in the supporting information. The comparison of the calculated and experimental ^1H NMR chemical shift values is shown in Appendix B in the supporting information. RMSD was calculated from the difference between the ^1H NMR of the experimental value and predicted value as outlined in (3). The variation of the experimental proton shifts with the corresponding predicted values using the four software was carried out for all the molecules. Figure 1 shows the results of the variation for molecule 1.

3.1.1. *Average RMSD*

RMSD is calculated as per (3) and the average RMSD was calculated from the RMSD of the 30 molecules and shown in the table below.

The results show the following trend in order of increasing RMSD, MestReNova < ACD Workbook Suite < ChemDraw ≪ NMRShiftDB.

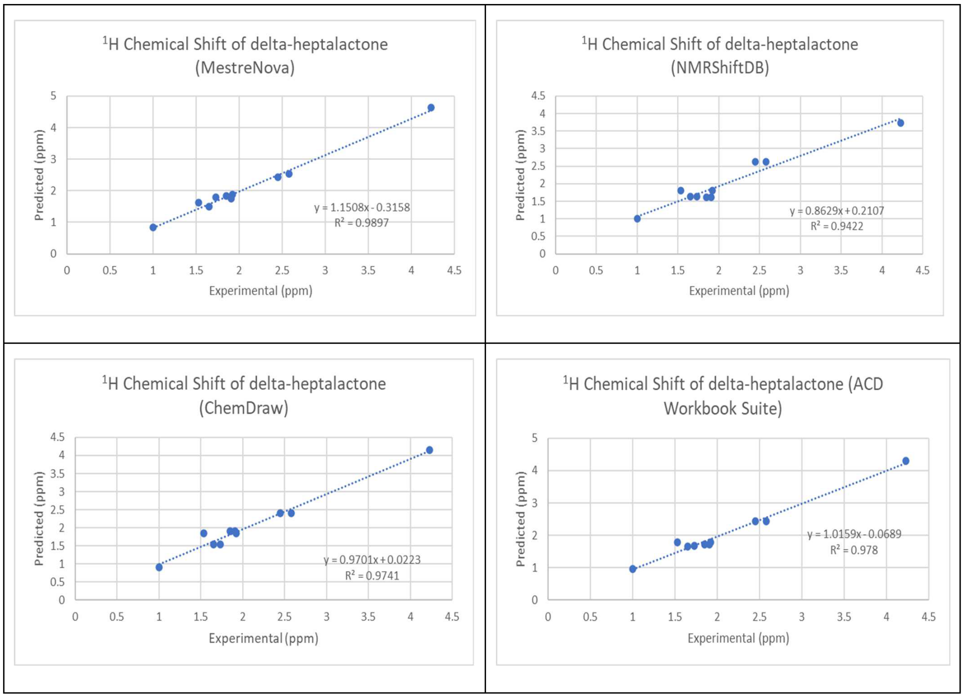


Figure 1. Variation of the experimental proton shifts with the corresponding predicted values for molecule 1.

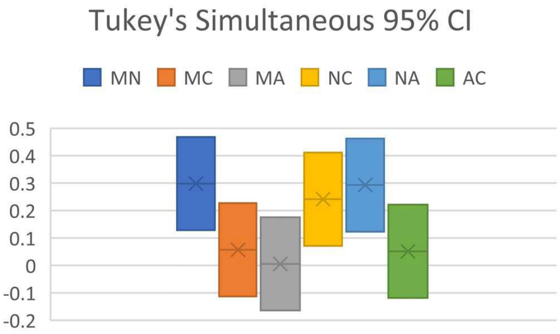


Figure 2. Tukey simultaneous 95% CI for predictor program pair combinations (M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

3.1.2. *ANOVA, Tukey's HSD and CI from RMSD of the 30 organic molecules*

To test for the significant difference in RMSD from Table 2, one-way ANOVA with a post hoc test and Tukey's HSD were carried out. The results are shown in Table 3a, 3b and 3c and Figure 2.

ANOVA of the dataset showed that $F = 9.38$, $F_{\text{crit}} = 2.68$, and $P = 0.0000133$. As $P < 0.05$ and $F > F_{\text{crit}}$, it suggests that H_0 is false, whereas H_1 is true: at least one of the predictor programs is significantly different in performance than the others. Tukey's HSD indicates that $Q_{\text{stat}} > Q_{\text{crit}}$ between NMRShiftDB paired with MestReNova, with ChemDraw, and with ACD Workbook Suite, affirming the conclusion reached by ANOVA. Results from Figure 2 shows the CI analysis where the CI between NMRShiftDB and all other programs does not intersect zero, indicating that there is significant difference between NMRShiftDB and all other programs, verifying the conclusion of Tukey's HSD.

The results obtained from RMSD lead to the conclusion that NMRShiftDB (with the highest average RMSD) is the weakest in ^{1}H NMR prediction whereas the remaining three predictors are equally good in performance.

3.2. *MAPE (%)*

MAPE (%) was calculated from the total percentage error as outlined in (2).

Table 2. RMSD of the 30 organic molecules

Molecule	RMSD of the 30 organic molecules			
	MestReNova	NMRShiftDB	ChemDraw	ACD Workbook Suite
1	0.158199558	0.223282556	0.139216019	0.131732304
2	0.132205018	0.507366156	0.429319714	0.231302399
3	0.250353084	0.412088178	0.46808653	0.248822226
4	0.250149132	0.313776616	0.275316716	0.28554893
5	0.153337714	0.398996127	0.221822411	0.134359423
6	0.097377429	0.101795696	0.117088156	0.244548564
7	0.157633637	0.408167969	0.200715085	0.254124164
8	0.991556128	0.991556128	0.092443376	0.023242681
9	0.184746313	0.254360374	0.10793146	0.363292444
10	0.064758292	0.326990409	0.099995454	0.283289925
11	0.169342355	0.290319077	0.110152167	0.46665628
12	0.164378167	0.415193493	0.182995777	0.1007084
13	0.153414797	1.611406249	0.219695471	0.192520908
14	0.231103061	0.455596998	0.436298207	0.186641367
15	0.042973247	0.291949824	0.029132456	0.121849087
16	0.075511589	0.335740972	0.260335797	0.157330573
17	0.116416398	0.392535207	0.43611097	0.116511325
18	0.348320398	0.440394255	0.244824631	0.44057576
19	0.329876837	0.996771425	0.285785361	0.382548036
20	0.093588272	0.500206957	0.34992764	0.281360978
21	0.275964529	0.553884464	0.426578308	0.378532554
22	0.209280016	0.295367356	0.331437211	0.299683062
23	0.090381414	0.394404361	0.15513478	0.332540373
24	0.617479738	0.883528232	0.656619684	0.683054603
25	0.058695187	0.316056957	0.078370913	0.051740941
26	0.459865431	0.548149289	0.468122923	0.40571084
27	0.624529823	1.068597211	0.513298159	0
28	0.480409201	1.732503785	0.229454491	0.260868654
29	0.181191762	0.407916547	0.979724943	0.127477984
30	0.17631676	0.418365391	0.502282391	0.317947165
Average	0.244645176	0.542908942	0.30160724	0.250150732

3.2.1. *Average MAPE (%)*

The average MAPE (%) was calculated from the MAPE (%) of the 30 molecules and is shown in Table 4.

The results show the following trend in order of increasing MAPE (%), MestReNova < ACD Workbook Suite < ChemDraw ≪ NMRShiftDB. Molecules such as compound 4 showed higher MAPE (%). The ^{1}H NMR of the protons in the chair conformation of its cyclohexane contributed to the high error. For molecule 22, the highest error was shown by the hydrogen in the cyclopentane. The ^{1}H NMR assumed a flat cyclopentane, however it could be either in

Table 3a. Single factor ANOVA from RMSD of 30 organic molecules

Source of variation	SS	df	MS	F	P value	F crit
Between groups	1.791134363	3	0.597044788	9.38209254	1.33255E-05	2.682809407
Within groups	7.38184953	116	0.063636634			
Total	9.172983893	119				

Table 3b. Tukey's HSD from RMSD of 30 organic molecules

Pair	Difference	Q stat	Significant statistical difference
MN	0.298263766	6.476010207	Yes
MC	0.056962064	1.236780827	No
MA	0.005505555	0.119538602	No
NC	0.241301702	5.23922938	Yes
NA	0.29275821	6.356471605	Yes
AC	0.051456508	1.117242225	No

(M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

Table 3c. Confidence interval (CI) from RMSD of 30 organic molecules

Pair	Difference	Upper CI limit	Lower CI limit
MN	0.298263766	0.468056457	0.128471075
MC	0.056962064	0.226754755	−0.112830627
MA	0.005505555	0.175298246	−0.164287135
NC	0.241301702	0.411094393	0.071509011
NA	0.29275821	0.462550901	0.122965519
AC	0.051456508	0.221249199	−0.118336182

(M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

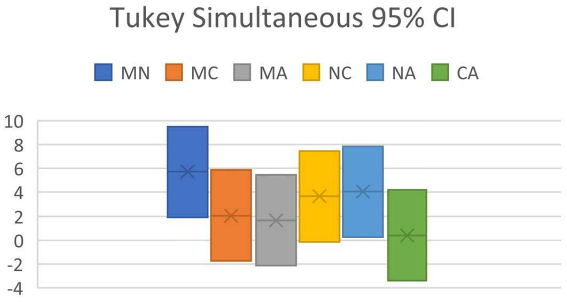


Figure 3. Tukey Simultaneous 95% CI for predictor program pair combinations (M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

envelope or half-chair conformation. Molecule 24 had various five- and six-membered cyclic moiety and its exact conformation was not considered in the ^{1}H NMR predictions. The same case was encountered with compound 26, where the six-membered ring protons gave the highest error.

3.2.2. *ANOVA, Tukey's HSD and CI from MAPE (%) of the 30 organic molecules*

To test for the significant difference in MAPE (%) from Table 4, one-way ANOVA with a post hoc test and Tukey's HSD were carried out. The results are shown in Table 5a, 5b and 5c and Figure 3.

ANOVA of dataset shows that $F = 5.49$, $F_{\text{crit}} = 2.68$, and $P = 0.00145$. As $P < 0.05$ and $F > F_{\text{crit}}$, it indicates that H_0 is false, whereas H_1 is true: at least one of the predictor programs is significantly different in performance from the others. In Tukey's HSD, it is found that $Q_{\text{stat}} > Q_{\text{crit}}$ between MestReNova and NMRShiftDB, and between NMRShiftDB and ACD Workbook Suite, in line with the conclusion reached by ANOVA.

Results in Figure 3 show the CI analysis where the CI between MestReNova and NMRShiftDB, and between NMRShiftDB and ACD Workbook Suite do not encompass zero, indicating that there exists significant difference between MestReNova and NMRShiftDB, and between NMRShiftDB and ACD, affirming the conclusion obtained by Tukey's HSD. The results obtained from MAPE lead to conclude that MestReNova and ACD Workbook Suite are equally good in performance, followed by ChemDraw and NMRShiftDB. This conclusion differs slightly from that obtained using RMSD.

Table 4. MAPE of the 30 organic molecules

Molecule	MAPE (%) of the 30 organic molecules			
	MestReNova	NMRShiftDB	ChemDraw	ACD Workbook Suite
1	5.717016795	7.757044772	6.374206787	5.705317882
2	3.457274581	14.90233757	15.12577338	7.372038802
3	2.754474665	4.060824356	4.75206928	3.015019914
4	15.63547599	18.69213871	16.15365045	14.68724293
5	4.134137032	7.480177279	5.017721695	3.509145976
6	1.804341875	3.473166633	2.645913839	7.528264736
7	1.58826661	7.374456369	3.521567751	7.299849584
8	8.584075843	7.179467635	1.53870372	0.493973606
9	3.654524363	6.098809222	2.134442416	13.57000892
10	1.384165294	8.092697791	3.249486222	9.203666198
11	3.128689071	6.653747635	2.516097881	15.15575568
12	1.951321346	7.3119789	2.52315348	3.550421853
13	7.753393404	40.0219553	6.563769996	9.194718558
14	2.979856941	5.289498235	5.780252463	2.960532135
15	1.420874825	7.109018275	1.044595353	3.749748505
16	1.321297802	8.26591674	7.02811375	2.461663019
17	2.546452891	5.690348764	5.488150679	3.175524462
18	5.10986636	10.06512895	3.039594832	5.356112132
19	5.342243544	14.86655201	3.271496429	5.810154517
20	3.952488954	13.91247433	15.2768481	7.191562469
21	5.288046347	13.0807625	15.36188933	8.205957251
22	12.87252284	17.02719402	19.34607702	14.74142521
23	2.836695567	7.768817027	4.26845463	7.62937135
24	15.28376466	21.82579088	16.67810021	19.63752167
25	0.970251774	5.573267219	1.279891869	0.884969588
26	11.32680729	11.99120799	10.9485264	8.640007363
27	7.233767583	18.62754286	17.64618799	0
28	6.135162462	12.26033072	4.988802026	4.26720172
29	2.196676326	4.903645145	2.937826634	1.526134792
30	2.549187235	5.02799848	6.227786948	4.33685596
Average	5.030437342	10.74614321	7.090971719	6.695338893

3.3. *Conclusion from RMSD and MAPE statistical studies*

The conclusion based on MAPE is that MestReNova and ACD Workbook Suite are equally good in performance, followed by ChemDraw and NMRShiftDB, because MAPE is more reliable as it normalizes our data over the experimental value to a range of 0% to 100%, as compared to RMSD which is unscaled. Based on Table 4, although MestReNova exhibits the lowest MAPE, it could be due to chance, as ANOVA and Tukey's HSD indicate that the difference in its mean is not significant as compared to ACD Workbook Suite.

Table 5a. Single factor ANOVA of RMSD of 30 organic molecules

Source of variation	SS	df	MS	F	P value	F crit
Between groups	522.096092	3	174.0320307	5.492488409	0.001454083	2.682809407
Within groups	3675.513547	116	31.68546162			
Total	4197.609639	119				

Table 5b. Tukey's HSD for MAPE (%) of the 30 organic molecules

Pair	Difference	Q stat	Significant statistical difference
MN	5.715705869	5.561609341	YES
MC	2.060534377	2.004981974	NO
MA	1.66490155	1.620015485	NO
NC	3.655171492	3.556627367	NO
NA	4.050804318	3.941593856	YES
CA	0.395632826	0.384966489	NO

(M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

Table 5c. Confidence interval (CI) for MAPE (%) of 30 organic molecules

Pair	Difference	Upper CI limit	Lower CI limit
MN	5.715705869	9.504451169	1.926960568
MC	2.060534377	5.849279677	−1.728210924
MA	1.66490155	5.453646851	−2.12384375
NC	3.655171492	7.443916793	−0.133573809
NA	4.050804318	7.839549619	0.262059018
CA	0.395632826	4.184378127	−3.393112474

(M = MestReNova, N = NMRShiftDB, C = ChemDraw, A = ACD Workbook Suite).

3.4. *Using NMR software to predict chemical shifts*

Using the best NMR predictors, the ^{1}H NMR chemical shifts of three published compounds, with 60 data points namely epitetrathiodiketopiperazine (referred to as Compound **31**), bis(p-fluorobenzyl)disulfide (Compound **32**), and tosylate (Compound **33**) [12], were predicted and compared with literature. The structural formulae of the compounds are shown in Figure 4.

The predicted ^{1}H NMR chemical shifts and experimental data are shown in Table 6.

3.4.1. *RMSD and MAPE (%) of three organic compounds from literature [12]*

Table 7 shows the average RMSD and MAPE (%) for MestReNova and ACD Workbook Suite calculated from the findings in Table 6.

Table 7 shows that MestReNova has a lower average MAPE and the same RMSD as compared to ACD Workbook Suite.

3.4.2. *T-test of RMSD and MAPE*

T-test was performed to determine the statistical difference between the average values obtained from MestReNova and ACD Workbook Suite. Statistical difference is significant when both following conditions are met: (a) TSTAT value is lesser than TCRIT value, (b) P ($T \leq t$) two-tail value is more than 0.05. When it is the case, the NMR prediction software with either the lower average RMSD or the lower average MAPE (%) has performed significantly better than the other. Based on the values obtained in both Table 8a and 8b, there was no instance in which both conditions proved that statistical difference was simultaneously reached.

The t-test showed that the difference between the average MAPE and RMSD of MestReNova and ACD Workbook Suite is not significant. Based on this result, we concluded that MestReNova and ACD Workbook Suite are equally good in terms of ^{1}H NMR prediction, even though the average MAPE of MestReNova is lower than that of ACD Workbook Suite, as the statistical tests have shown that there is no significant difference between their average MAPEs.

4. Conclusion

In contrast to NMR spectrometers, NMR prediction programs are certainly more cost-effective, versa-

Table 6. The predicted ^{1}H NMR chemical shifts and experimental data

Hydrogen position	Experimental (ppm)	Predicted (ppm)	
	Literature	MestReNova	ACD Workbook Suite
^{1}H NMR chemical shift of compound 31			
SO$_2$Ph-o-H	7.96	7.95	7.859
SO$_2$Ph-p-H	7.54	7.71	7.495
SO$_2$Ph-m-H	7.435	7.58	7.455
8	7.435	7.17	6.997
7	7.235	7.36	7.207
6	7.05	7.04	6.848
5	6.99	7.2	7.71
2	6.89	6.03	6.492
2′	6.84	7.12	7.095
3′	6.7	6.83	6.64
5′	3.76	3.8	3.767
12a	3.27	3.15	3.716
12b	3.1	2.97	3.587
17	3.05	3.14	2.845
18	1.99	1.87	1.873
^{1}H NMR chemical shift of compound 32			
8	7.64	7.17	6.992
SO$_2$Ph-o-H	7.59	7.71	7.859
3″	7.365	7.34	7.317
SO$_2$Ph-p-H	7.365	7.95	7.495
7	7.365	7.36	7.207
SO$_2$Ph-m-H	7.16	7.58	7.455
5	7.16	7.2	7.72
6	7.16	7.04	6.848
3‴	7.08	7.34	7.317
4″	7.04	7.09	7.049
4‴	6.91	7.09	7.044
2′	6.74	7.12	7.105
3′	6.63	6.83	6.64
2	6.59	5.98	6.352
15	4.91	5.6	5.192
1″a	4.21	3.99	4.073
1‴a	4.01	3.96	4.035
1″b	3.83	3.96	4.073
1‴b	3.79	3.94	4.035
5′	3.77	3.8	3.767
12a	3.55	2.96	3.693
17	3.12	3.1	3.056
12b	3.06	2.95	2.571

(continued on next page)

Table 6. (continued)

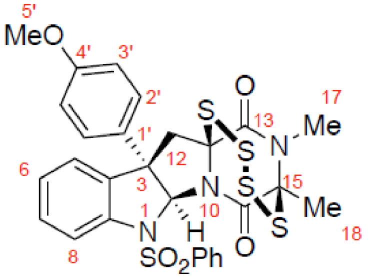
Hydrogen position	Experimental (ppm)	Predicted (ppm)	
	Literature	MestReNova	ACD Workbook Suite
^{1}H NMR chemical shift of compound 33			
9′	7.77	7.62	7.7
8	7.61	7.19	6.997
SO_2Ph-o-H	7.47	7.71	7.753
SO_2Ph-p-H	7.34	7.95	7.495
7	7.275	7.36	7.217
10′	7.275	7.46	7.34
5	7.105	7.23	7.389
6	7.105	7.04	6.856
SO_2Ph-m-H	7.105	7.58	7.455
2′	6.66	7.11	7.115
3′	6.54	6.83	6.809
2	6.15	5.86	6.052
11	4.39	4.55	4.256
5′	4.25	4.11	4.007
15	4.04	4.43	3.66
7′	3.95	4.17	4.125
12a	3.13	2.54	3.035
12b	2.88	2.46	2.572
17	2.88	2.91	2.87
12′	2.4	2.39	2.447
6′	2.13	2.16	2.159
18	1.58	1.31	0.822

Table 7. Average RMSD and MAPE (%)

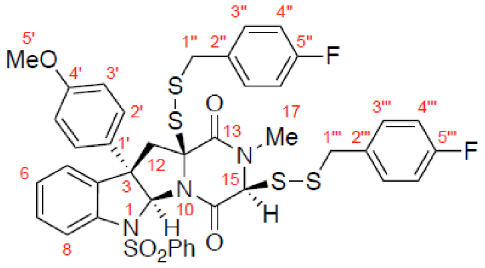
Molecule	MestReNova	ACD Workbook Suite
	RMSD	
Compound **1**	0.267129806	0.314466214
Compound **2**	0.320154514	0.278211993
Compound **3**	0.311245664	0.300011515
Average	0.299509995	0.297563241
	MAPE (%)	
Compound **1**	3.279491594	4.940945337
Compound **2**	4.287435252	4.002110785
Compound **3**	5.613675005	5.885956871
Average	4.39353395	4.943004331

tile, and time-saving, as they are easily accessible and acquirable through the internet. In addition, NMR software allows different types of NMR predictions based on a variety of interchangeable solvents and frequencies. The ability to employ high frequencies in NMR prediction allows the chemical shift data to be more pronounced while saving cost on acquiring high-end spectroscopic instruments. NMR prediction programs can rapidly generate chemical shift data with high accuracy using well-studied and reliable computational methods and include other functions such as peak processor, multiplet analysis, structure elucidation programs, and embedded chemical shift databases. The ability to rapidly generate highly accurate chemical shift data without using any spectroscopic instruments makes NMR prediction programs an invaluable tool for scientists.

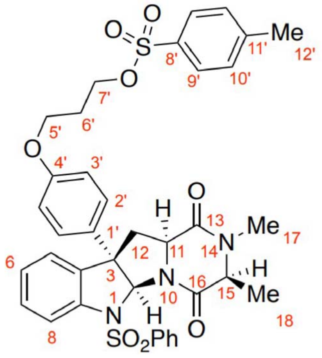
The data obtained from this project suggest that among the four ^{1}H NMR software, MestReNova and



Compound **31**



Compound **32**



Compound **33**

Figure 4. The structural formula of epitetrathiodiketopiperazine (Compound **31**), bis(*p*-fluorobenzyl)disulfide (Compound **32**), and tosylate (Compound **3**), with their atoms labelled.

Table 8a. *t*-test: RMSD of 3 organic compounds from literature

	MestReNova	ACD Workbook Suite
Mean	0.299509995	0.297563241
Variance	0.000806199	0.000333088
Observations	3	3
Pearson correlation	−0.885835578	
Hypothesized mean difference	0	
d*f*	2	
t Stat	0.074338735	
$P(T \leqq t)$ one tail	0.473753524	
t Critical one tail	2.91998558	
$P(T \leqq t)$ two tail	0.947507049	
t Critical two tail	4.30265273	

Table 8b. *t*-test: MAPE (%) of three organic compounds from literature

	MestReNova	ACD Workbook Suite
Mean	4.39353395	4.943004331
Variance	1.370545749	0.887222199
Observations	3	3
Pearson correlation	0.567987829	
Hypothesized mean difference	0	
d*f*	2	
t Stat	−0.949287589	
$P(T \leqq t)$ one tail	0.221334666	
t Critical one tail	2.91998558	
$P(T \leqq t)$ two tail	0.442669333	
t Critical two tail	4.30265273	

ACD Workbook Suite provide equally accurate ^{1}H NMR predictions, with NMRShiftDB and ChemDraw predictions having equally less accuracy than the previous two programs. The equal level of accuracy between MestReNova and ACD Workbook Suite, and between ChemDraw and NMRShiftDB is due to the inability of the data collected to prove statistical significance between the two programs within the respective pairings, and thus we cannot definitively confirm that one program is better or worse than the other within each pairing. The results of this study can prove extremely useful in deciding which software to be prioritized in order to obtain more accurate ^{1}H NMR predictions, encouraging the use of NMR software that provide accurate predictions, and discouraging the use of programs that do not.

Conflicts of interest

Authors have no conflict of interest to declare.

Acknowledgements

The authors are grateful to ACD/Labs and IT Tech Research (M) Sdn. Bhd. for providing free access to ACD software for 30 days.

Supplementary data

Supporting information for this article is available on the journal's website under https://doi.org/10.5802/crchim.156 or from the author.

References

[1] C. Cobas, "Ensemble NMR prediction", 2019, https://resources.mestrelab.com/ensemble-nmr-prediction, (accessed March 2021).

[2] MODGRAPH, "HOSE code and substituent chemical shift prediction", http://www.modgraph.co.uk/product_nmr_substit.htm, (accessed March 2021).

[3] D. C. Burns, E. P. Mazzola, W. F. Reynolds, *Nat. Product Rep.*, 2019, **36**, 919-933.

[4] C. Cobas, *Magn. Reson. Chem.*, 2020, **58**, 512-519.

[5] J. Meiler, W. Maier, M. Will, R. Meusinger, *J. Magn. Reson.*, 2002, **157**, 242-252.

[6] J. Aires-de Sousa, M. C. Hemmer, J. Gasteiger, *Anal. Chem.*, 2001, **74**, 80-90.

[7] M. E. Elyashberg, A. J. Williams, G. E. Martin, *Prog. Nucl. Magn. Reson. Spectrosc.*, 2008, **53**, 1-104.

[8] J. Gordon, "NMR and IR spectra predictions in ChemDraw and Chem3D, ChemDraw and Chem3D have several tools for predicting NMR and IR spectra", http://cbn.cambridgesoft.com/Articles/articlesPrinterVersion.aspx?articleID=644, (accessed February 2021).

[9] C. Steinbeck, S. Krause, S. Kuhn, *J. Chem. Inf. Comput. Sci.*, 2003, **43**, 1733-1739.

[10] A. Bahl, "MAPE v/s MAE % v/s RMSE", 2019, https://agrimabahl.medium.com/mape-v-s-mae-v-s-rmse-3e358fd58f65, (accessed March 2021).

[11] J. Frost, "Using post hoc tests with ANOVA", 2019, https://statisticsbyjim.com/anova/post-hoc-tests-anova, (accessed March 2021).

[12] C. R. Olsson, J. N. Payatte, J. H. Cheah, M. Movassaghi, *J. Org. Chem.*, 2020, **85**, 4648-4662.