Comparative nuclear magnetic resonance study of *Natrum muriaticum* LM1 produced in-house and obtained from commercial sources respectively.

By

Garth Meredith Miller

Dissertation submitted in partial compliance with the requirements of the Master's Degree in Technology: Homoeopathy in the Faculty of Health Sciences at the Durban University of Technology.

I, Garth Meredith Miller, do declare that this dissertation is representative of my own work, both in conception and execution.

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Signature of student **Date of signature**

APPROVED FOR FINAL SUBMISSION

_____ _____

Signature of Supervisor **Date of signature**
Dr I. Botha
DTech: Homoeopathy

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Signature of Co-Supervisor **Date of signature**
Prof A.H.A. Ross
DTech: Homoeopathy

DEDICATION

To my cherished wife and soulmate, Lynne:

Without you, this endeavour would not have been possible, in all things you remain as my similimum.

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ABSTRACT

Introduction

Hahnemann was adamant that his methodology for LM1 prescription be reproduced exactly, in order to develop the latent medicinal properties of the crude substance reliably (Barthel 1991). Yet quality assurance practices during manufacturing are currently aligned to Good Manufacturing Process, in adherence to Pharmacopeia which permit substantial deviation from the strict quantities and practices (Kayne 2006) prescribed in Hahnemann's 6th edition *Organon* (Hahnemann and O'Reilly 2001). Often the equipment, methods and practices favour expedience or utilise new technologies, under an assumption that the remedy produced is unaffected, since the mechanisms underlying the action of homoeopathic remedies are not understood or determined (Barthel 1991).

Aim

The primary purpose of this study was to compare and evaluate the nuclear magnetic resonance (NMR) spectra of *Natrum muriaticum* LM1, produced using strict adherence to original Hahnemannian methodology, to samples sourced from a representative variety of sources available to local homoeopathic practitioners, in the medicinal prescription form patients may receive.

Additionally, the researcher set out to confirm that these *Natrum muriaticum* LM1 samples could be shown to produce distinct NMR spectra when compared to a Lactose LM1 control.

The final objective was to ascertain whether two samples produced with the same strict adherence to Hahnemannian methodology, from same source materials would yield similar NMR Spectra, distinct from a Lactose LM1 control sample.

Methodology

The researcher produced an in-house *Natrum muriaticum* LM1 (0/1) sample in accordance with aphorism 272 of the 6th edition *Organon (Hahnemann and O'Reilly 2001).* Similarly, a second sample was produced by the DUT Homoeopharmaceutics Senior Lecturer, within the same ambient conditions and protocol. A third sample was produced by a local make-to-order (MTO) company, using the same sodium chloride crude substance sample utilised in the production of the first two samples. These three samples emulate the make-to-order (MTO) options available to homoeopathic practitioners.

A fourth sample was obtained ex-stock from highly regarded local supplier (Homoeopathix Trading Company (South Africa)), while the fifth and sixth samples were procured ex-stock from highly regarded suppliers abroad, Helios (United Kingdom) and Roy & Co. (India) respectively. These samples

represented the make-to-stock (MTS) sourcing options available to homoeopathic community.

The researcher transported the samples to Chemistry Department of the University of Stellenbosch Chemistry Department in Cape Town. Any external influences such as vibration, changes in temperature, electromagnetic disturbances, heat or strong light were avoided or minimised as far as was practical.

The six LM1 potency samples were prepared immediately prior to NMR analysis from their respective source materials to a 20% alcohol concentration, as could be done in practice for the patient prescription where the alcohol content would serve as a preservative.

Four controls comprised the seventh, eighth, ninth and tenth samples, for comparative analysis and to confirm sample homogeneity.

- Lactose (LM1)
- Source lactose in solution
- Water used in preparation of NMR samples
- Ethanol solvent

The ten samples were assigned a random reference number by Dr Jaco Brand (Nuclear Magnetic Resonance Unit Manager), to remove bias during NMR analysis. Five samples were drawn from each of the ten randomised

samples and labelled, while deuterated dimethyl sulfoxide-d6 (DSMO) contained within a separate capillary tube served as an external lock and reference solvent.

The researcher then carried out the analysis under qualified supervision to ensure best-practice was applied throughout. The resulting data (FID) was processed to derive the chemical shift and relative integration values, which were captured into a Microsoft® Excel 2010 spreadsheet to calculate the relative integration values of each sample run. Statistical analysis was performed in GNU PSPP version 085, the data was analysed using descriptive statistics and the non-parametric tests, Kruskal-Wallis and Mann-Whitney U-test (due to small data size and nature of distribution) at a statistical significance interval of $\alpha = 0.05$.

Results

Natrum muriaticum LM1 samples sourced from a variety of make-to-order (MTO) and make-to-stock manufacturing environments produced almost universally distinct NMR spectra profiles in terms of chemical shifts and relative integration values of the CH_2, CH_3 and H_2O signals, at an alcohol concentration of 20%, when compared to a Lactose LM1 control sample.

When the NMR spectra of make-to-order (MTO) and make-to-stock (MTS) remedies were analysed and compared to each other in terms of chemical shifts and relative integration values of the CH_2, CH_3 and H_2O signals, they

were for the most part statistically distinct, but some anomalous results emerged.

There was no statically significant difference in NMR spectra in terms of chemical shift, between the two samples produced in the same environment using the methodology to closely aligned to *Organon* (Hahnemann and O'Reilly 2001), yet in terms of relative integration values, they were statistically distinct.

Conclusion

The low alcohol percentage associated with the medicinal dosage of LM1 prepared for analysis failed to produce an OH peak on NMR spectra. In addition, it is not possible to identify the variables or indeed account for the nature of NMR spectra that are produced after sample analysis.

This study concludes that while NMR was able to discern physico-chemical distinction between LM1 potency *Natrum muriaticum* remedies to control conclusively, this does not translate to practical application of NMR for quality control or remedy comparison for LM potencies at alcohol content levels, associated with medicinal prescription.

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ABBREVIATIONS

ANOVA	Analysis of Variance
B_o	External magnetic field
CH	Centesimal (Hahnemannian)
CH_2	Methylene group
CH_3	Methyl group
DUT	Durban University of Technology
FID	Free induction decay
FT	Fourier Transform
g	Gram
ggt	Drop
GMP	Good Manufacturing Practice
H_2O	Water
Hz	Hertz
J	Coupling constants
ml	Millilitre
MOW	Memory of water
MTO	Make-to-order
MTS	Make-to-stock
Nat-m.	Natrum muriaticum
NMR	Nuclear Magnetic Resonance
OH	Hydroxyl group
Ppm	Parts per million
RF	Radio frequency
T_1, T_2	Relaxation times in NMR
Δ	Delta
µl	Microlitre
UV	Ultra violet

DEFINITION OF TERMS

Agitation

The application of a disturbing force to a liquid.

Aphorism

Concise statement of a scientific principle, typically by a classical author.

Analysis of Variance

A method of statistical analysis in which the differences between group means are analysed.

Analysis of Variance (ANOVA)

A parametric statistical method used to analyse data testing for significant differences in the means.

Avogadro's number

The number of units in a mole of any substance equal to 6.0225×10^{23}.

Batch

A specific quantity of medicine, which is uniform in characteristic and quantity within specified limits, and is produced at the same time according to a single specified manufacturing procedure.

Centesimal

The most commonly used dilution scale used in homoeopathic pharmacy. It represents a dilution level of 1:100 so that each succeeding potency thus contains one hundredth part of the preceding potency. The centesimal scale is denoted by CH or C.

Chemical shift

In NMR spectroscopy, it indicates the resonance frequency of nuclei in an external electromagnetic field relative to a reference standard such as Trimethylsilane. It is measured in parts per million (ppm) of the operating frequency of the spectrometer.

Cavitation

The formation of vapour cavities or voids within a liquid subjected to varying pressures. Such cavities implode when the pressure of the liquid increases and release shockwaves into the surrounding liquid.

Ellipsometry

Optical technique which is very sensitive to any change of optical response related to incident radiation interacting with the material investigated. Consequently this technology is able to investigate dielectric properties (complex refractive index or dielectric function) of thin films.

Fifty-millesimal potency scale

This homoeopathic scale dilutes substances at a ratio of 1:50 000, thought to have a deeper and more gentle action compared to other potency scales.

Hahnemannian

Pertaining to philosophy, principles and method prescribed by Samuel Hahnemann, founder of homoeopathy.

Integration

The relative intensity of individual NMR peaks. It is indicative of the relative number of hydrogens present at each signal.

ISO

International Organization for Standardization

Larmor frequency

The characteristic frequency of the precessional motion of a charged nucleus when placed in an external magnetic field during NMR experimentation. The Larmor frequency is specific to each nucleus.

LM potency scale

This homoeopathic scale dilutes substances at a ratio of 1:50 000, thought to have a deeper and gentler action compared to other potency scales.

Materia Medica

Encyclopaedia of the substances which have been used to prepare homeopathic medicines. They list the materials along with details of the provings which establish the symptoms and conditions for which they are claimed to be suitable.

Mean (x)

A measure of central tendency defined as the sum of the values in the sample divided by the sample size.

Multiplets

Multiple peaks seen together on NMR spectrum.

Natrum muriaticum

Homoeopathic preparation of sodium chloride (table salt).

NMR-spectroscopy

An analytical method that employs an applied magnetic field to measure its effect on the protons present within organic compounds and thereby reveal information about the structure of such compounds. The interaction between the applied magnetic field and the compound is recorded as a series of peaks known as an NMR spectrum.

Pharmacopoeia

Reference book describing the preparation and use of medicines.

Physical structure

Three-dimensional arrangement of atoms and molecules in a compound or mixture.

Polycrest

Commonly used homeopathic medicines are called polychrests or polycrest remedies.

Posology

Study and application of dose of medicine, in homoeopathy dose incorporates potency, quantity and form of medicine, but also repetition.

Potency

An altered state of material substance used as a homoeopathic medicine. The potency level is indicative of the deconcentration level of that remedy.

Potentisation

A process used in the preparation of homoeopathic medicines to raise potency levels. The remedy undergoes serial dilution and agitation, typically by succussion and/or trituration, in a specified ratio.

Precession

Refers to the change in orientation of a body's rotational axis. In the context of NMR, usually in response to an external magnetic force.

Proving

The scientific method through which the therapeutic remedy picture of a homoeopathic remedy is obtained and referenced. The remedy is administered to healthy participants in order to ascertain the type of morbid symptoms it is able to produce. This information can then be collated and recorded in the *Materia Medica*.

Quinquagenimillesimal potency scale (Q-potency)

This homoeopathic scale dilutes substances at a ratio of 1:50 000, thought to have a deeper and gentler action compared to other potency scales.

Relative integration values

The integration of an area beneath a particular peak, evaluated relative to the total area beneath all the peaks on an NMR graph. It is calculated by dividing the integration values of each peak by the sum of all the integration values of the run.

Repertorisation

Repertorisation is not only a mechanical process of counting rubrics and totalling marks obtained by a medicine, it also includes the logical steps to reach the repertory proper and finally differentiating the remedies with the help of *Materia Medica*

Remedy

A medicine produced in accordance with the homoeopathic pharmaceutical process and homoeopathic philosophy.

Solvatochromic dyes

Solvatochromic dyes to measure solvent parameters to explain solubility phenomena and predict suitable solvents for particular uses.

Spectrum (NMR)

A graph depicting, by means of a series of peaks, the resonant frequencies at which protons of different types of hydrogen atoms of a compound

absorb electromagnetic radiation. Ethanol for example produces four peaks: H_2O, OH, CH_2, and CH_3.

Standard deviation

A quantity expressing the extent to which the members of a group differ from the mean value for the group.

Succussion

Agitation of a liquid homoeopathic remedy that employs firm swift shakes in a vertical plane terminating with a jolt. Often this action is performed by hand, pounding the remedy container repeatedly against an elastic surface.

Thermoluminescence

Thermally stimulated luminescence is a well-known phenomenon amongst the thermally stimulated processes, where studied material is “activated” at low-temperature, usually by radiant energy. When the irradiated material is warmed up, the heating trigger release of accumulated energy and a characteristic glow is emitted.

Trituration

The process of grinding lactose and a medicinal starting substance together by means of a mortar and pestle for a prolonged period of time in order to reduce the particle size of the substance and render insoluble substances soluble.

T-tests

In statistics, an analytical method that is employed to determine the differences between the means of two groups.

CHAPTER 1 - INTRODUCTION

Building on the current body of knowledge which has tended to focus on the theoretical variables of potentisation in isolation (Ross 1997; Power 1999; Lyell 2004; Botha 2005), this research evaluated whether subtle variances between the variables inherent during remedy preparation methodology aggregate to a distinguishable difference in NMR spectra.

This has both direct and practical implications for homoeopathic practitioners as remedies are routinely procured from manufacturers where production efficiency, manufacturing best practice and profit motives may compromise the strict adherence to Hahnemannian methods of production, but the impact of this variation on remedy produced is not known or understood (Barthel 1991). Rather, it is assumed these variations do not alter the remedy or its effects as elicited during the original proving, where the remedy was likely produced under an entirely distinct methodology and procedure. Significant structural differences between samples used during proving and those produced subsequently could signify changes in medicinal application, as was believed to be the case.

If an equivalent potency (LM1) of the same remedy (*Natrum muriaticum*) is shown to produce different NMR spectrum profiles, then it is probable that the potentisation variables (succussion, dilution, trituration) which have been shown in other studies to impact on NMR spectrum profiles, have aggregated to affect this change, due to a lack of standardisation between the production processes and/or the variables inherent to remedy production environments. This could have far-reaching implications for homoeopathic proving's (for symptom determination) and repertorisation (determining applicable remedy) where lack of standardisation would be fundamental to these practices.

Significant variation between NMR profiles would imply that structural variances have occurred at an atomic level which is believed to be the mechanism for transfer of energetic memory for curative action (Resch and

Gutmann 1987), so the action of the remedy itself may be affected (Hahnemann and O'Reilly 2001). This would have direct implications for homoeopaths and the homoeopathic community at large (Vithoulkas 1998; Bloch and Lewis 2003), especially regarding the need for more rigorous standardisation throughout *Materia Medica* for remedy preparation, likely extending beyond the LM potency scale chosen here. Additionally, homoeopathic practitioner's may find this insight invaluable in selection of their suppliers, while manufacturers could factor this impact (if found to be significant) in their production methodologies, with ultimate goal being the improved care of homoeopathic patients through reproducible results.

Toward the end of his life Hahnemann developed the LM (quinquagenimillesimal / Fifty-millesimal) potency scale which triturates the source remedy in mortar and pestle with lactose to 3CH (centesimal scale), then has a dilution scale of 1/50 000 and involves 100 succussions between each two-step liquid and potency stage derived initially from the LM mother tincture (Barthel 1991; DeSchepper 2001; Hahnemann and O'Reilly 2001). In the 6th Edition of the *Organon* aphorism 270 prescribes the process for LM potency manufacture, but much latitude is applied by companies and individuals involved in production and use of these remedies, while many misconceptions related to LM potencies exist today (DeSchepper 2006). There is very little knowledge related to the impact of this variation on remedy properties, so it is hoped this research will provide some insight to this aspect of homoeopathic practices (Milgrom *et al.* 2001).

1.1 The Aims of the Study

The aim of this study was to compare and evaluate the nuclear magnetic resonance (NMR) spectra of LM1 homoeopathic medicinal potency samples of *Natrum muriaticum* comprising:

- Two sourced from suppliers readily available to local practitioners

- Two conveniently sourced imported samples
- Two produced in-house as per GHP

NMR spectra were compared in terms of chemical shifts and relative integration values of the CH_2, CH_3, H_2O and OH signals, in order to determine differences in their respective physical structure.

1.2 The Objectives of the Study

1.2.1 The First Objective

To compare and evaluate the nuclear magnetic resonance (NMR) spectra of the LM1 homoeopathic medicinal potency samples of *Natrum muriaticum* to the lactose LM1 (control) sample, which has been potentised to LM1 potency as per GHP, with respect to the chemical shifts and relative integration values of the CH_2, CH_3, H_2O and OH signals, so as to determine differences in physical structure.

1.2.2 The Second Objective

To compare and evaluate the nuclear magnetic resonance (NMR) spectra of the six sources of LM1 homoeopathic potency samples of *Natrum muriaticum* with one another, in terms of chemical shifts and relative integration values of the CH_2, CH_3, H_2O and OH signals, in order to determine if statistical differences can be distinguished between their respective physical structures using this technology.

1.2.3 The Third Objective

To ascertain whether two samples produced with the same strict adherence to Hahnemannian methodology, from same source materials and environmental conditions, would yield similar NMR Spectra, distinct from a Lactose LM1 control sample.

1.3 The Hypotheses

1.3.1 The First Hypothesis

It was hypothesised that preparation practises are an important determinant of remedy characteristics as stated by Hahnemann (Barthel 1991; Hahnemann and O'Reilly 2001), so where variation in manufacturing a given homoeopathic remedy exists, evidence of differences will be reflected in results obtained from NMR investigation.

1.3.2 The Second Hypothesis

It was hypothesised that the *Natrum muriaticum* LM1 remedy samples will be distinctive in comparison of samples to the LM1 lactose control.

1.3.3 The Third Hypothesis

It was hypothesised that two *Natrum muriaticum* LM1 remedy samples produced with the same strict adherence to Hahnemannian methodology, from same source materials and environmental conditions, would yield similar

NMR Spectra, distinct from NMR spectra obtained from a Lactose LM1 control sample.

1.4 Assumptions

It was assumed that remedies were received in the state intended during their production and that they were not subjected to any unforeseen environmental exposure that may have altered their nature during storage, transport or processing.

At LM1, the extent of dilution is approximately 5.5CH, so just below the Avogadro's number (6CH) (Morgan and Helios Pharmacy 2014), so physical properties detected and observed using NMR are likely due to atomic alignment, representing a relevant and meaningful reflection of the intrinsic nature of samples evaluated.

It is assumed that the powerful electromagnetic field which is inherent to NMR spectrometry technology, will effectively demonstrate:

- Physical differences between the molecular properties of potentised samples and when compared to controls, which are relevant to action of homoeopathic remedies
- Homoeopathic samples will not be altered by the powerful electromagnetic fields that are inherent to the NMR technology,, despite the commonly held belief that homoeopathic remedies are sensitive to electromagnetic fields.

1.5 Delimitations:

- This research was not intended to identify the specific nature or cause related to the results materialised.
- There is no intention implicit within this research to derive a conclusion as to the therapeutic implications due to any physical characteristics found to exist.
- The extent of research confined to the single remedy *Natrum muriaticum* and only to medicinal potency LM1, no extrapolation beyond that scope is proposed or implied.
- The research only investigated the physical properties observable in the NMR spectra and did not seek to explain the samples in terms of other visualisation technology or analytical methods
- There is no intention to assess the nature, accuracy or suitability of any other method of detecting physical attributes of homoeopathic remedies.
- While reference is made to the possible application of NMR technology in quality control and standardisation of homoeopathic remedies produced, there is no intention to quantify this technologies suitability in these applications.

- There is no intention, explicit or implied to criticize, compare suppliers or assert that any conclusion can be derived related to suitability, quality or therapeutic value from the NMR analysis data derived. This investigation is limited to merely processing remedies through NMR to discern spectra differences and highlight material and methodology as potential variables for the results attained.

CHAPTER 2 - REVIEW OF RELATED LITERATURE

2.1 Introduction

More than two centuries ago, Samuel Hahnemann began documenting his journey toward improving the quality of medical care and developing an entirely new medical system, known as homoeopathy. His initial dilution of medicinal substances was motivated by this desire to avoid the side effects of crude dosages, but it was his observations, experimentation and clinical practice with micro-dilutions that led to his discovery of potentisation (Banerjee 2006).

He continually improved remedy manufacturing and prescription practices to ensure consistency and improve patient outcomes, through extensive experimentation with the variables related to potentisation (dilution scales, succussion, and trituration) and posology (potency, form, quantity and repetition) then analysed his meticulous notes to identify further opportunities for improvement (Whitney 2010).

With his introduction of LM potencies, which represented a culmination of more than half a century of his experience (Barthel 1993), Hahnemann introduced extensive changes in philosophy, manufacturing and posology, detailed throughout his 6th edition of the *Organon* (Barthel 1991; Hahnemann and O'Reilly 2001), which was later published posthumously in 1921. Hahnemann was convinced LM potency taken in liquid form, offered profound

benefits and was preferable to his decimal and centesimal potencies (Hahnemann and O'Reilly 2001).

However, today there is still no quantitative way to reliably ensure a remedy received was produced using the exact same methodology employed, or that it is therapeutically equivalent to the potentised substance produced for the initial proving, or indeed from one batch to the next. Today, many manufacturing practices for remedies produced, bear little resemblance to the standards and philosophy Hahnemann believed were vital and to which he demanded strict adherence to his methods detailed in the *Organon* (Hahnemann and O'Reilly 2001).

Barthel (1991) believes that only strict adherence to these instructions will guarantee consistent results and is critical of pharmacopoeia that use relative quantities and the latitude afforded to methods of succussion and trituration. The two *Natrum muriaticum* samples produced at DUT at LM1 potency for analysis and evaluation, emulate these instructions closely to determine whether NMR profiles obtained will indeed support this assertion, when compared to remedies produced from the same source substance where methodology and environment differs.

Homoeopathic dispensaries incur a significant cost and remedies are believed to directly impact patient outcomes, so practitioners source from the suppliers they believe produce superior quality remedies. However, even where Good Manufacturing Practice (GMP) and Quality Assurance (QA) are implemented

by the supplier, a multitude of variables exist where the impact on the remedy is unknown. It is likely that until the mechanisms underlying homoeopathic potentisation are better understood and can be subjected to objective and accurate measurement, this impact on posology is at best conjecture (Botha 2014). So what should the practitioner consider when procuring remedies until that becomes a reality?

Unfortunately, the ability of remedies to retain information related to the original substance is currently not understood, although several theories and hypotheses exist regarding possible mechanisms for the 'memory of water' and encouraging research into at ultra-diluted solutions is ongoing. Yet given these methodology inconsistencies during the production of the samples used in this field of research, this may be a significant factor accounting for the lack of consistency in much of this research undertaken. It is hoped that by comparing remedies of known origin to commercially manufactured counterparts in this study, this variable may be better elucidated and understood.

Several promising technologies today may be able to reliably detect and reflect the physical properties unique to homoeopathic preparations being used in research. This researcher exposed the remedy *Natrum muriaticum* (0/1) obtained from a collection of diverse sources, then applied the exact same method during the last stage of preparation of patient prescript (LM1) as is routine in practice (DeSchepper 2006). The last stage of preparation involves conversion of 0/1 (granule), into liquid to produce the patient

prescription (at 20% alcohol), this stage was kept as consistent as was practically possible. (This method is detailed in appendix D - LM1 sample for analysis). It was hoped that the resulting NMR evaluation yields spectra that enables insight into the impact of methodology as a variable.

Nuclear Magnetic Resonance (NMR) as used in this investigation is considered among the most promising technologies available, as it is has a considerable body of research where it has been able to detect changes physico-chemical structure of homeopathic dilutions (Kayne 2006; Botha 2009). A change in structure at this level of dilution, beyond Avogadro's number, would not be attributed to a physical agent so consequently could support the concept of 'water memory', even if the exact mechanism is yet to be determined (Weingaertner 1992).

This approach has been successfully applied to demonstrate changes in chemical shift and relative integration values within extreme dilution levels, when variables related to potentisation and manufacturing methodology have been compared to controls. For example, utilising *Natrum muriaticum* (same source substance for this study), NMR analysis revealed distinct chemical shift and relative integration spectra when the degree of succussion was evaluated, which suggests that potentisation methodology is indeed fundamental to the resulting physicochemical nature inherent to homoeopathic remedies (Lyell 2004)

Since the results of this type of analysis are promising, NMR could evolve as a practical technology to derive reference points to validate the consistency of physical structure within potentised remedies and/or serve as a motivation to identify technologies better suited for this task. This could manifest as a practical and important quality assurance mechanism that practitioners could use to ensure they receive a consistent and clinically predictable remedy.

2.2 Samuel Hahnemann - Evolution of Homoeopathic Remedies

Christian Friedrich Samuel Hahnemann (1755 – 1843) is the only human being known to have conceived an entirely unique system of medicine, distinct from the prevailing paradigm and successfully develop that vision into a practical and efficacious system within his own lifetime (DeSchepper 2006). He qualified as a medical practitioner at the University of Erlangen in 1779, but he soon became disillusioned by the prevailing medical practices, especially bloodletting and routine administration of toxic substances in crude dosages, among them mercury, arsenic and opium. He was condemned by the profession for his advocacy of better diet, hygiene practices, more exercise and rest in preference to accepted practices, in preference to established 'real' medical practices which he openly criticised for the adverse effects it inflicted on patients (Haehl 1922; DeSchepper 2006).

Consequently, he left practice in 1781, to further his studies in chemistry and during this period utilized his fluency in several languages to translate medical texts to supplement his income (DeSchepper 2006). So it was that in 1791, at

46 years of age that Hahnemann experienced a turning point, while translating 'Cullen's *Materia Medica*' into German (Haehl 1922). Cullen was a Scottish physician and teacher (Passmore 2001), this was his seminal work where he attributed the anti-malarial potential of Cinchona bark (contains anthraquinone, active ingredient in quinine, at that time a primary treatment for malaria for more than two centuries) to its astringent and bitter qualities.

Hahnemann was intrigued, since several compounds he knew to have similar properties, were ineffectual in this application and this is evidenced by a footnote he added in the reference document (Taylor 1998). This prompted him to consume Cinchona bark (aka Peruvian Bark) himself as an experiment to observe its effect on a healthy individual (Vermeulen 2004; Owen 2007).

Hahnemann soon began to illicit symptoms characteristic of malarial fever, which led to his profound insight that substances with curative properties, illicit symptoms characteristic of those diseases where it has application. Cinchona bark has proved to be an important homoeopathic remedy, China (Vermeulen 2001). However, this revelation motivated Hahnemann to extensively review medical literature and ultimately derive the 'First Law of Homoeopathy', also known as the 'Law of Similars' (*Similia similibus curentur*), best understood as "like cures like" (Bloch and Lewis 2003; DeSchepper 2006). This fundamental premise is already documented in his '*Organon* of the rational art of healing' (Hahnemann 1810).

This quote from James Tyler Kent highlights the significance of this principle in the practice of Homoeopathy clearly and succinctly: "*Hahnemann distinctly declares that the phenomena of cure depends entirely upon fixed law, the 'law of similars', or the law that governs homeopathy*" (Kent 2000: 90). For practitioners, this principle manifests practically as a pursuit for the patient's 'similimum' during a consultation. A homoeopathic remedy which matches the specific expression of the patients' symptoms, when administered to a healthy individuals during homoeopathic 'provings', could be considered a similimum for that individual, it's this endeavour that remains as Hahnemann's true legacy (Whitney 2010).

Hahnemann continued to formulate, refine and improve his philosophies and manufacturing practices, based on his own experiences in clinical practice, objective results of experimentation, insights and understanding. This progression is meticulously recorded as evidenced in his surviving journal entries, publications, case histories, correspondences and across all six editions of the *Organon*, revealing his dedication, perseverance and give insight to his remarkable vision (Schmidt 2010), his meticulous nature evident in his five subsequent *Organon* editions, that serve as a reflection of the evolution of his understanding, methodology and application (Owen 2007; Whitney 2010).

Hahnemann's experience of prescription in high doses, the medical paradigm of that period, appears to have motivated his experimentation with increasingly higher levels of dilution of the source crude substances to

mitigate the associated undesirable side effects (DeSchepper 2006). Indeed his knowledge and experience using crude doses allopathically, form the foundation of homoeopathic application (Master 2012). Initially, he prescribed very low doses of crude substances loosely resembling homoeopathic principles today, which he first officially published details of in 1796, in his "*Essay on a New Principle for Ascertaining the Curative Powers of Drugs, and Some Examinations of Previous Principles"* (Taylor 1998).

Through Hahnemann's notes and published works we can infer that succussion, was preceded by an intention to thoroughly incorporate substances by mixing and shaking between successive serial dilutions. The extent of dilution was progressively increased and extended to include serial dilution stages by 1799, detailed in a publication he titled "*Cure and Prevention of Scarlet Fever*" published in 1801, which elaborated on the use of highly diluted Belladonna (then termed a "weak solution") in successful treatment and for prophylaxis during the Scarlet Fever epidemic of 1800 (Taylor 1998; DeSchepper 2006).

He made this illuminating entry into his personal journal in 1825:

"*In the preparation of homoeopathic attenuations a small portion of medicine is not merely added to an enormous quantity of non-medicinal fluid, or only slightly mingled with it but by the prolonged succussions and trituration, there ensues not only the most intimate mixture, but at the same time and this is the most important circumstance, there ensues such a great and hither to unknown and undreamt of change, by the development and liberation of the*

dynamic powers of the medicinal substance so treated, so as to excite astonishment" (Banerjee 2006: 336)

At this point Hahnemann appears to be contemplating the energetic nature of remedies liberated through dilution and the role of shear-forces during succussion as a "potentisation" and "dynamization" of the crude substance.

Hahnemann's experimentation with trituration was catalysed after reading about this practice being used by Arabian physicians on gold. He began experimenting with this method and first documented it in the *Materia Medica Pura* (Volume 4) in 1818 (Barthel 1991). By 1828 he confirmed that with this process where metals were triturated in three stages, each stage of one hour duration, were rendered soluble in water (3CH trituration), in his publication of Chronic Diseases (Volumes 1-3) (Barthel 1991). Trituration would evolve, becoming an integral component in the production of LM potency remedies detailed in the 6th edition of the *Organon* (aphorisms 246-248, 270-271, 278) (Hahnemann and O'Reilly 2001; Jütte 2007), but only published 82 years posthumously in 1921 (Master 2012).

In contrast to the evolving and experiential nature of his remedies, Hahnemann remained consistent and precise regarding our obligation as physicians within his visionary medical system of homoeopathy, as declared in aphorism 1 across all the six editions of the *Organon*, *"The physician's highest and only calling is to make the sick healthy, to cure, as it is called*" (Hahnemann and O'Reilly 2001).

He probably died with the conviction that LM remedies aligned best with his vision for best possible cure, as detailed in all editions of the *Organon* of Medicine in aphorism 2, promoting "*rapid, gentle and permanent cure*" (Hahnemann and O'Reilly 2001). He considered LM potencies unlikely to cause homoeopathic aggravations despite having a more powerful action and liquid administration employed as superior (Castro 1996; De Schepper 1999).

2.3 LM Potency in Homoeopathy

2.3.1 Development of LM Potency Scale

De Schepper (1999) illustrates the radical evolution LM potencies represented by highlighting that in order to incorporate this philosophy and manufacture resulted in an effective overhaul of the *Organon* (5th edition), requiring 60 new aphorisms , 49 amendments and 40 aphorisms deletions for the 6th edition of the *Organon* (De Schepper 1999). Hahnemann referred to his 50 millesimal potency (1:50 000) homeopathic remedies as the 'LM potency scale', derived from the Roman numerals where 'L' represents 50 and 'M', 1000. Today it's also common practice for this potency to be referred to as the 'Q-potency', derived from the Latin root for 50 000 (quinquagintamilia) (Jütte 2007).

Hahnemann attributed the high level of dilution combined with intensive potentisation (100 succussions) at each stage, combined with the relative low degree of overall succussion for the deep, yet gentle action associated with LM potencies with reduced potential toward homoeopathic aggravations he observed clinically (De Schepper 2005; Banerjee 2006; Kayne 2006). By

introducing prescription of LM potencies as a liquid form, now based on a linear scale this simplifies patient management and enables subtle differentiation of each dose (Owen 2007).

2.3.2 Manufacturing Logic and Methodology

By 1821, in *Materia Medica Pura* (volume 6) Hahnemann was able to clearly describe how to properly perform manual succussion, '*bring down ten times, using the full strength of the arm*' to potentise remedies between serial dilutions (Barthel 1991). He continued to experiment and refine succussion and trituration and develop his understanding of these variables within the context of progressive serial dilution. By the time succussion becomes inherent to methodology for LM potencies, this combination was the end result of more than half a decade of extensive experimentation and refinement (Barthel 1991; DeSchepper 2001).

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Hahnemann and LM Methodology

The methodology meticulously detailed by Hahnemann in aphorism 270, of his 6th edition *Organon* (Barthel 1991; Hahnemann and O'Reilly 2001; Jütte 2007) is the realisation of his life's work, so with deep respect is used as a basis for the following outline of his method (see Hahnemann and LM Methodology).

In summary, After C3 trituration and preparation of a 1:50 000 Mother tincture, Hahnemann serially diluted centesimally (1:100) with each successive potency receiving 100 succussions, before impregnate granules at a ratio of

1:500, which yields the resulting 1:50 000 dilution desired. Hahnemann was meticulous in his use of measurement units prevalent in chemistry and pharmacology at the time, was specific regarding method and materials and adamant that strict adherence to this process was required for results (Hahnemann and O'Reilly 2001).

Barthel (1991) advocates for strict adherence to all aspects of Hahnemann's methodology, criticising German Pharmacopoeia for its use of relative values and guidelines related to succussion. He contends that manufacturing cannot simply be ramped-up to industrial volumes, and mechanised succussion and trituration practices, without impacting on the properties of the remedies produced (Barthel 1991, 1993).

He also reminds us that Hahnemann expected physicians to prepare their own remedies in aphorism 265 (Hahnemann and O'Reilly 2001), even considering that rest periods would enable practitioners to employ the same energy through trituration and incorporating them into his method (Owen 2007) and described in Hahnemann and LM Methodology. Research into the impact of producing in bulk is lacking in literature since sample preparation during research is usually on a small scale. This motivated this study to procure and evaluate commercially produced remedies ex-stock, from sources locally sought after and perceived as producing high quality remedies.

2.4 Homoeopathic Quality Assurance in Practice

Compliance to Hahnemann's Mandate must surely be our default priority in the absence of definitive quantitative evaluation of properties inherent and unique between homoeopathic remedies. Today, few homoeopaths are able to produce their own remedies due to the labour-intensive nature, legislation, expense and complexity of sourcing source substances and applying quality assurance measures to ensure remedies are safe and efficacious.

Consequently there is a significant cost associated with stocking their dispensaries adequately and physicians believe doing so assures remedies of a high quality and is in their patients' best interest. It is likely that until the mechanisms underlying homoeopathic potentisation are fully understood and are subject to objective and accurate measurement the impact on posology is at best conjecture (Botha 2014). What should the practitioner consider when procuring remedies until that becomes a reality?

"*It is infinitely easier to contradict than to investigate...*" - *Samuel Hahnemann (Haehl 1922: 91).* Progressively, regulatory bodies and manufacturers have deviated from Hahnemann's mandate and process of remedy evolution. This progression in an apparent absence of a comprehensive understanding of underlying dynamics or modern technology to determine the best course, so based primarily on consensus and expedience, not on the relentless investigation and disciplined process advocated (indeed practiced) by the founder (Kayne 2006). Other possibilities accounting for discrepancies to

Hahnemann's method include errors in translation, misunderstandings and the LM potencies methodology being described alongside methods for other potencies with the pharmacopoeia (Barthel 1991).

Hahnemann's approach however, was not based on self-interest or expedience. Despite criticism, professional exclusion and the peronal and financial hardships he suffered as a result, he persevered and refined his methodology based on meticulous experimentation, motivated by his commitment to patient care (Haehl 1922; DeSchepper 2006). His methodology evolved continually, but his conviction regarding our responsibility to our patients remained constant. "*I am not ashamed to confess that I did not know yesterday what experience might teach me today. Experience and scribbling are two very different things" - Samuel Hahnemann (Haehl 1922: 80).*

Do we best honour Hahnemann's legacy through the strict adherence he demanded, as suggested by Barthel (1991), or instead facilitate manufacturing efficiencies and inclusion of advancement in technologies? Until we have the means to quantitatively determine the impact of the variables within remedies and on patient care – perhaps more caution, even regarding minor deviations from his methodology would be advisable.

No reliable representation of the physical properties of the potentised remedy (in high dilution) is currently available to ensure consistency and efficacy of the remedy received (Sahani 2007). So in practice, Good Manufacturing

Practice (GMP) is primarily orientated to isolation of source substance, purity, safety, sampling, inspection of inputs and various production-related activities (Kayne 2006). Ultimately suppliers and practitioners assume that if these criteria are met, the remedy will perform to match the accumulative features described in the relevant materia media, and homoeopathic repertories.

Nevertheless, deviations from the original methodology have evolved and are now represented in the various pharmacopeia, inherent to GMP reference by companies and laboratories. Also, manufacturers routinely adopt practices that are based on economic and logistical considerations, presuming no impact (indeed changes may currently have no measurable impact) on the final remedy they supply (Power 1999). This includes novel methods of succussion like sonication, which employ '*Intense ultrasonic waves traveling through liquids*' (Suslick 1989: 80) for their effect. Improbable potentisation practices like radionics may show statistically significant impact in research (Allsopp 2010; Kleingeld 2015), but certainly we cannot yet be sure this consequently implies an equivalent therapeutic action to original proving substances. All attempts to locate research that would support this assertion comprehensively have not been successful.

The routine focus of quality control is based on isolating a remedies 'active ingredients' or profile, according to existing industry norms using technology like thin film chromatography, then assuming that the medicinal nature of the substance is limited to, or predominately due to that isolated component and not the substance in its natural combined state, which is the prevailing

paradigm in allopathic medicine. Davey (1992) clarifies the purpose of quality assurance as aligning with other medical disciplines, so accurate standardisation of therapeutic preparations is then considered prerequisite to efficacy and safety.

It is not entirely clear if these quality assurance practices are actually applicable, necessary or beneficial in preparation of homoeopathic remedies, or indeed if they serve the patients best interests as intended, again, because there is no objectively quantitative way to achieve this to satisfaction of the parties involved. Ultimately, aligning to best practice within an allopathic paradigm usually prevents practitioners from producing their own remedies, through legislation or cost burden of compliance, yet this was advocated as being desirable by Hahnemann (Hahnemann and O'Reilly 2001). Also, the presumption that a source substance has an isolated 'active ingredient' which describes the therapeutic profile of that substance, which may be composed of many more compounds simply deemed irrelevant as there nature, role and quantity assign them this status, in homoeopathic terms may be presumptive. This was a motivating factor in the selection of *Natrum muriaticum* (Nat-m) for NMR analysis, since this variable could be eliminated.

Over time, weights and measures have evolved and been standardised, but now differ from the original proportions, scale and process used in Hahnemann's instructions (Hahnemann and O'Reilly 2001). These updated measures, standard and values are however inherent to the pharmacopoeia

referenced by suppliers and practitioners, so how can we be sure that there has been no impact on the nature of these remedies? (Botha 2014)

Botha (2014) explains that the apothecaries' system of measures (based on fluid ounce) was used by physicians during Hahnemann's lifetime, but differed depending on country due to differing standards. However, in France the non-metric measurements were replaced by Metric (SI units) in 1840. In Table 2-1, Botha (2014) clearly highlights distinct differences which have emerged in the German Homoeopathic Pharmacopoeia (GHP) and Homoeopathic Pharmacopoeia of the United States (HPUS) representations of Hahnemann's original methodology, detailed in aphorism 270 (§270) of 6th edition *Organon* (Hahnemann and O'Reilly 2001). His meticulous instructions were based on a measurement system in use at that time, which was subsequently improved, but the conversion into the new system adopted does not appear to have been accurately transcribed.

- Trituration originally referred to troy grains (0.053g substance and 5.5g substrate) as he was using the French Apothecary system in §270 (Botha 2014)
- GHP retains integrity of methodology but is based on ratios
- HPUS also uses ratios but amends proportions to 1:99, eliminates the three-fold addition of diluent and doesn't specify trituration time. The new reference standards clearly deviate from the original and assume that the energy imparted is adequate irrespective of the volume processed.

	§270 and footnotes (Hahnemann 1999; O'Rielly 2010)	**GHP** (Benyunes 2005)	**HPUS** (Homœopathic Pharmacopoeia Convention of the United States 2013)
Trituration of the 1C	One **grain** triturated for three hours with three times one hundred **grains** sugar of milk: 1. 1/3 of 100 **grains** sugar of milk with one **grain** powdered drug 2. Triturated rather strongly 6 to 7 minutes 3. Scraped from mortar and pestle 3 to 4 minutes 4. Repeat steps 2 and 3 5. Add second third of sugar of milk 6. Repeat steps 2 to 4 7. Add last third 8. Triturate as before for 6-7 minutes with careful scraping	<u>Method 6</u> One **part** triturated for three hours with three times one hundred parts lactose monohydrate: 1. 1/3 of 100 **parts** lactose monohydrate with one part powdered drug 2. Triturated by hand for 6 minutes 3. Scraped from mortar and pestle 4 minutes 4. Repeat steps 2 and 3 5. Add second third of lactose monohydrate 6. Repeat steps 2, 3 and 4 7. Add last third lactose monohydrate 8. Repeat steps 2, 3 and 4	<u>Method 33</u> Prepare by triturating one (1) **part** of the medicinal raw material with ninety-nine (99) **parts** of lactose monohydrate. Triturate for a sufficient time (to ensure that a homogenous mass is prepared.). The result is the 1C trituration.
Trituration of the 2C	Repeat for 2CH (1/10000)	Repeat for C2	One (1) **part** of the 1C trituration is triturated with ninety-nine (99) **parts** of lactose monohydrate. Triturate for a sufficient time. The result is the 2C trituration.
Trituration of the 3C	Repeat for 3CH (1/1000000)	Repeat for C3	One (1) **part** of the 2C trituration is triturated with ninety-nine (99) **parts** of lactose monohydrate. Triturate for a sufficient time. The result is the 3C trituration.

Table 2-1: LM Potency Guidelines (Botha 2014)

Liquid potentisation in use today has deviated even more from aphorism 270 (§270) of 6th edition *Organon* (Hahnemann and O'Reilly 2001).

- LM mother tincture strictly following Hahnemann's instructions would entail 0.053 grams of 3C triturate dissolved in 500 drops (26.5 grams) of an alcohol and distilled water mixture.
- GHP instructs 60mg of C3 triturate dissolved in 20ml of 15% (m/m) alcohol.
- HPUS ratios this step as one part (by weight) of 3C trituration dissolved in five hundred parts (by volume) of alcohol / distilled water mixture.

Impregnation of dry granules also differs considerably:

- While Hahnemann prepared dry LM potency by saturating 26.5g (5 grains) of granules with a single drop of the liquid LM potency
- GHP refers to moistening 100g of granules.
- HPUS has five hundred parts of no10 pellets (by weight), exposed to one part (by volume) of the LM potency.

	§270 and footnotes (Hahnemann 1999; O'Rielly 2010)	**GHP** (Benyunes 2005)	**HPUS** (Homœopathic Pharmacopoeia Convention of the United States 2013)
Q mother tincture preparation	1 **grain** dissolved in 500 **drops** of a mixture of one part alcohol (Hahnemann 1999) (brandy wine = 90° alcohol (O'Rielly 2010)) to four parts distilled water	Method 17 **60mg** dissolved in **20ml** of 15% (m/m) alcohol = 500 drops	Methods 32-33 To one (1) **part** (by **weight**) of the 3C trituration, add five hundred (500) **parts** (by **volume**) of a mixture composed of one (1) part 95% (v/v) alcohol and four (4) parts water. Dissolve the 3C trituration completely.
Q1 - liquid	One **drop** and 100 **drops** of pure alcohol (Hahnemann 1999) (good wine spirit = 95° grain alcohol (O'Rielly 2010)) (vial 2/3 full) and 100 succussions by hand against a hard but elastic body	1 **drop** to **2.5ml** 86% (m/m) alcohol = 100 drops and succuss 100 times by hand	To one hundred (100) **parts** (by **volume**) of 95% v/v alcohol add one (1) **part** of the liquid prepared in step 31.1.1. Succuss (see §28). The result is the 1LM attenuation.
Q1 – dry (0/1)	500 **granules**, where 100 **granules** weighing 1 **grain** is to be moistened with one **drop** dynamised alcohol in thimble (glass, porcelain or silver) with a small opening at the bottom, stirred and emptied onto blotting paper	100**g** size1 pillules (50 000) is to be moisten with solution	To five hundred (500) **parts (by weight**) of #10 pellets, add one (1) **part (by volume**) of the 1LM attenuation. Shake to ensure all the pellets are impregnated.
Q2 - liquid	One **globule** dissolved in one **drop** of water and 100 **drops** of good alcohol added and dynamised with 100 powerful strokes	1 **pillule** dissolved in 1 **drop** purified water and **2.5ml** 86% (m/m) alcohol added and succuss 100 times by hand	Dissolve one (1) **part (by weight)** of impregnated pellets from step 32.1.3 in one hundred (100) **parts (by volume)** of 95% v/v alcohol. Succuss (see §28). The result is the 2LM attenuation.
Q2– dry (0/2)	500 **granules**, where 100 **granules** weighing 1 **grain** is to be moistened with one **drop** dynamised alcohol in thimble (glass, porcelain or silver) with a small opening at the bottom, stirred and emptied onto blotting paper	100**g** size1 pillules (50 000) is to be moisten with solution	To five hundred (500) **parts (by weight**) of #10 pellets, add one (1) **part (by volume**) of the 2LM attenuation. Shake to ensure all the pellets are impregnated

Table 2-2: LM Potency Guidelines (Botha 2014)

Practitioners remain largely unaware that remedies are produced using variable methodology, or indeed that the pharmacopoeia are distinct from each other and that neither replicate Hahnemann's method exactly.

Today, more than two centuries since inception, there is still no scientifically verifiable explanation detailing the action of homoeopathic remedies or substantiate the homoeopathy's core tenants of 'infinitesimal dose' and 'law of similars'. Consequently the medicinal benefits patients realise in practice will continue to be attributed to the 'placebo effect' and 'psychosomatic' influence, despite a multitude of meta-analytical studies indicating otherwise. Yet despite considerable efforts to discredit homoeopathy, it prevails primarily due to the therapeutic benefits patients derive from it, without the side effects and consequences inherent to the predominant medical paradigm (Castro 1990; Ullman 1991).

Central to the debate regarding scientific method and burden of proof is the necessity of 'double-blind' verification in clinical trials, rather than the effectiveness of treatment (Almirantis 2013). Homeopathic remedies are initially taken by healthy individuals, during a 'proving' process, where symptoms expressed are documented and later prescribed on an individualised basis (peculiar and characteristic expression within the patient, rather than on emphasis on specific diagnosis) to each patient where it represents a similimum (Griffith and Castro 2010). The practical implication is that each patient with a specific condition (diagnosis) may be prescribed a unique remedy and/or potency in practice, so double-blind studies are seldom useful even though many studies of this nature have nevertheless had positive outcomes (Ullman 1991; DeSchepper 2006).

2.4.1 Homoeopathic Principles and Avogadro's Number

To account for the therapeutic benefits of potentisation, homoeopathic remedies are believed to imprint and retain an energetic signature of the source substance, which has been sourced, dissolved, succussed and exposed to serial dilution during preparation. Homoeopathic research into possible mechanisms to substantiate this 'memory of water' continues and several approaches investigating this phenomena show some promise. According to Griffith, homoeopathy was not founded on intellectualisation, rather it was aligned with the 'immutable laws of nature' so although simple to explain, are difficult to repeat exactly through statistical analysis and experimentation (Griffith and Castro 2010). This is compounded when poor quality research is the root cause of failure or poor perception of homoeopathic research, but in high quality studies it has been shown that positive outcomes far outweigh the negatives (Walach *et al.* 2005).

Hahnemann discovered that diluting a substance reduced not only its inherent toxicity but also the therapeutic benefit, but discovered that inducing kinetic energy at each stage of serial dilution (potentisation) resulted in progressively more profound therapeutic impact (Vithoulkas 1998). This principle known as the '*Law of Infinitesimal Dose*' which manifests when a homoeopathic remedy (potentised) is prescribed such that it conforms to the 'Law of Similars' and the curative potential homoeopathic potency then increases in direct ratio to that extent of dilution with succussion (Lilley 2012).

Unfortunately remedies produced and prescribed at extreme levels of dilution present particular challenges. Within the laws of chemistry there is a limit to which a substance can be diluted, so (in theory) beyond that level there will be none of original substance remaining. This limit relates to Avogadro's number (6.02254×10^{23}), a constant for the number of units in one mole of any substance (equal to a substance molecular weight in grams). In allopathic clinical pharmacology physiological and biochemical effects are believed to diminish with reducing dose of active 'ingredient' and certainly no therapeutic impact would be anticipated at dilutions well before Avogadro's number became a consideration.

Consequently, diluting beyond that level implies none of that substance should be present (Banerjee 2006), so any medicinal action is then implausible within existing chemistry or physics paradigm, which would necessitate another explanation be determined to account for the therapeutic action (Waisse 2012). Homoeopathic remedies, which employ serial dilution, reach this level at 24 iterations on the decimal potencies (24X) and only 12 on the centesimal potencies (12C) and at about four on the 50 millesimal scale (LM4) but homoeopathic prescriptions routinely far exceed this this dilution level.

The Arndt-Schulz Law is a hypothesis based on observations made by H. R. Arndt and Professor H. Schulz on the apparent action of substances at different concentration levels of dosages. They found that when considering living systems in general, low doses stimulate physiological activity, while medium doses inhibit it and high doses are detrimental to it (Kayne 2006).

Although this principle has been demonstrated in pharmacology as the 'Biphasal Paradoxical Effect' and has synergy in homoeopathy's 'Law of minimum dose' and 'Law of similars', it's application in dilutions beyond Avogadro's number are not clear and believed to rely on presence of the crude substance (Teixeira 2014).

A recent study was however able to demonstrate nanoparticles present in extreme dilutions (6CH, 30CH, 200CH, 1M, 10M and 50M) of Ferrum metallicum using high-resolution transmission electron microscopy (HRTEM) and energy dispersive spectroscopy (EDS), since these highly sophisticated methods are able to identify and locate nanoparticles in many homeopathic drug solutions. Rajendran concludes that the chaos created within the liquid retains nanoparticles well past Avogadro's number (Rajendran 2015). In addition, as these samples were drawn from centre of the liquid column, another hypothesis suggesting nanoparticles congregate to the surface at high dilution levels between stages of serial dilution accounting for their presence at extreme dilutions (Chikramane *et al.* 2010), may need revision.

2.5 Nuclear Magnetic Resonance (NMR)

2.5.1 Introduction

"*The faint radiofrequency signals detected in nuclear magnetic resonance (NMR) spectroscopy provide a window into the structure and dynamics of atoms in solids, liquids, and gases. No other experimental technique comes close to the range of atomic-level information that NMR can provide*" (Benesi 2015).

When samples are placed in a magnetic field and subjected to radiofrequency (RF) radiation (energy) at specific frequencies, nuclei in the sample can absorb the energy i.e. experience Nuclear Magnetic Resonance (James 1998). Nuclear Magnetic Resonance analyses the samples within a strong and stable magnetic field, the sample is also subjected to an electromagnetic field at specific frequency, able to interact with the nuclear dipole (Hardy 2011).

Any change in the properties of atomic structure or relative positioning (atomic spin) is then detected on the NMR spectrum and represented by 'Chemical shift' (resonant frequency of nuclei when exposed to an electromagnetic field) and 'Relative Integration' (integral area calculated under the graph peaks, of chemical shift). In this way NMR studies highlight differences in magnetic properties of protons (nuclei) within molecular compounds within a sample (Hofmeyr 2004), so allow insight into the composition and character which are inherent to that orientation, implicit to that specific molecular structure.

The ability to interpret the output NMR spectra enables determination of molecule structure, but is based on physical phenomenon that within a controlled magnetic field (B_o) energy required for nuclear spin flip occurs at magnetic resonance which can be measured.

2.5.2 Component Parts

NMR analysis requires highly specialised and costly equipment known as a NMR spectrometer (Figure 1), which comprises:

- A powerful superconducting magnet, configured to accommodate a probe though its core to precisely orientate the sample for analysis within the most stable realm of its magnetic field, and ideally oriented to the radio frequency coils (Benesi 2015).
- A Helmholtz coil is located in the sample probe and is used to produce accurate and powerful radio frequency signals to the sample, which are subsequently received from the sample by the detector, in order to record and analyse the absorption characteristics at given frequencies.
- A computer controls the hardware, using the proprietary VnmrJ Software interface (Schreiber 2007), configures the test parameters, feeds and ejects the sample and processes the NMR signals used to produce the spectra.

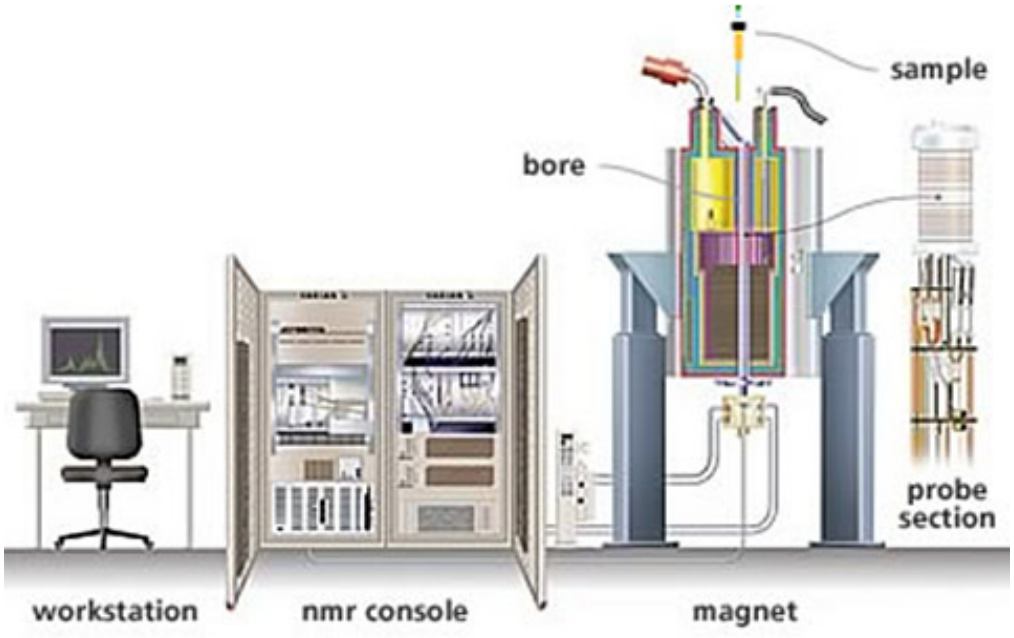

Figure 1: Modern NMR Spectrometer Components (Agilent, 2013)

The temperature of the superconducting wire coil for the superconducting magnet is well below the critical superconducting temperature, through immersion in liquid Helium. This ensures there is no resistance to current flow in the coil responsible for sustaining the uniform magnetic field inherent to NMR for extended periods with external power required.

The sample is pneumatically fed at atmospheric pressure from the top of the feed tube, positioned to align within the probe's radiofrequency coil, which was inserted in the bottom of the tube. Within the tube the sample is spun to average out any field inconsistencies present.

This research made use of the Agilent Inova 600 MHz NMR spectrometer at Stellenbosch University, Central Analytical Facilities Unit, which is able to resonate with the proton nucleus (1H) spin at a frequency of 600 MHz, due to its magnet field strength of 14.1 Tesla. The desired current and magnetic field through the sample is optimised with small adjustments to the shims (electromagnets located in the "shim stack" of the probe), such that magnetic lines of force are virtually parallel and of equal magnitude, with this static magnetic field (B_0) (Benesi 2015). A radio wave is introduced from the Helmholtz coil perpendicular the sample tube (B_1) and receives the signal emitted from the sample (see Figure 2).

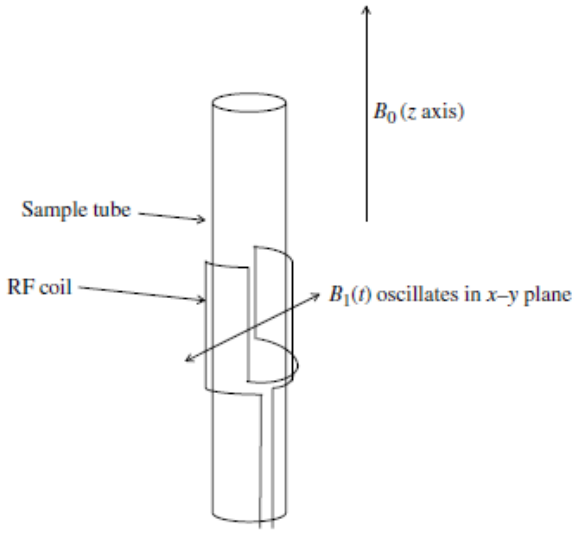

Figure 2: Helmholtz RF Coil (Benesi 2015)

2.5.3 Nuclear Spin

Magnetic spin is a property of nuclei having an odd atomic number or mass resulting in an associated magnetic moment, which results since the nucleus is a charged particle in rotation around its axis. Fortunately ^{1}H has a nuclear spin of ½, so can be imagined to resemble a miniature bar magnet with a magnetic field, called a magnetic moment (μ). (Teng 2013; Bakhmutov 2015). The mass relative to the spinning nucleus forms a angular momentum of this spin (μ), while the rotating charge generates a magnetic dipole moment (see Figure 3), proportional to and parallel to the vector of this spin angular momentum (Benesi 2015). Spinning particles are known to 'precess' around an applied external magnetic field (B_0) at an angular frequency known as the Larmor frequency.

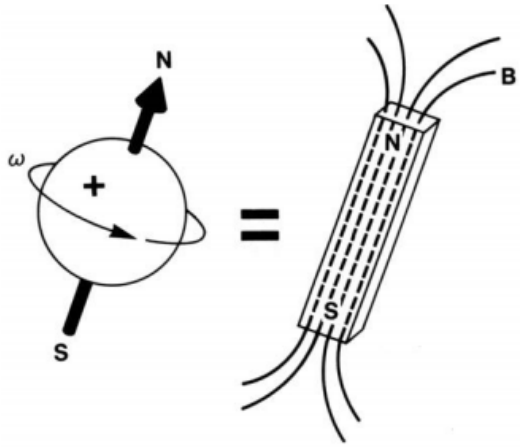

Figure 3: Charged nucleus with angular momentum, acts like a bar magnet

When placed in a strong external electromagnetic field (B_0), such as is produced by the superconducting magnet of a NMR spectroscopy device, these nuclei orientate themselves such that they either conform or oppose the field direction (B_0), so adopt their 'spin state'. This phenomenon is known as the 'Zeeman Effect' where difference between 'spin-states' is proportional to the strength of the magnetic field applied (B_0) (Teng 2013). The majority of nuclei will correspond with the applied field since this represents the lower energy state.

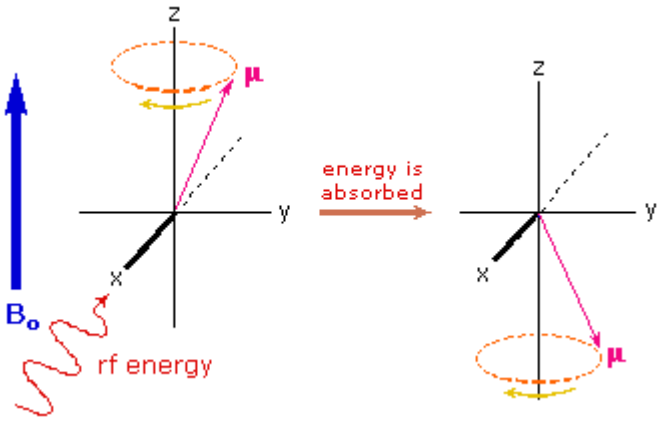

Figure 4: Excitation at Larmor frequency (Reusch 2013)

When an external radio frequency force (f_1) exerted on the nuclei matches the energy differential (Larmor frequency) of the precession nuclei, these nuclei 'resonate' absorbing enough energy to re-orientate and align with the higher energy state (Figure 3). The more powerful the applied external magnetic field (B_0), the larger the energy differential generated between these orientations, so the higher the resonant frequency required to elicit the change to higher energy orientation.

When radio frequency energy (f_1) is no longer applied, the nuclei discharge energy as they return to previous lower energy orientations within the constant magnetic field (B_o) of the superconducting magnet, signal is received by the probe to be amplified and recorded (Figure 4). The resonant frequency at which a proton will absorb energy is determined by its interaction with other atoms and elections, predominantly the nature of the electromagnetic field in which it exists. This is what enables specific and reproducible deduction of proton identity possible.

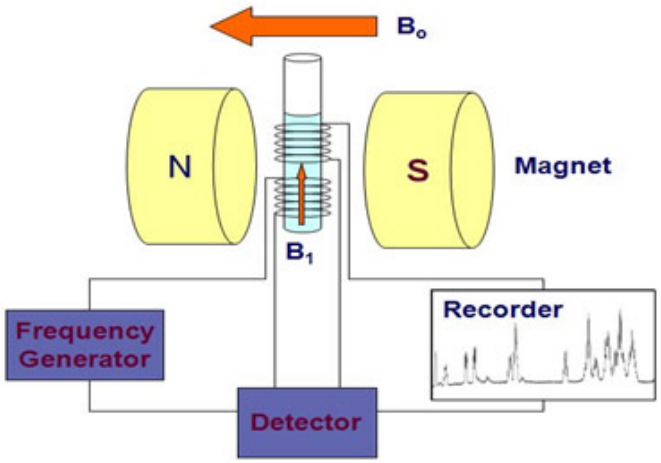

Figure 5: Simplified Schematic Illustrating NMR (Agilent 2013)

Nuclei which display resonance only at higher frequency are consequently recorded on the left and are considered poorly shielded. Conversely, as the extent of shielding increases (with increased electron density), the susceptibility of nuclei to the given field strength reduces, lowering the difference between 'spin states' and so the resonance frequency is also lowered.

2.5.4 Chemical Shift (δ)

Chemical shift is a measure of the relative resonance energy required by a nucleus in its given environment to elicit a flip in the spin of nuclei at lower energy state, for a given field strength (B_o), given the symbol delta (δ) and observed as a peak at given ppm on NMR Spectra. These NMR spectra are referenced across a horizontal axis where frequency increases from right to left, measured in parts per million (ppm) in relation to standard reference, like tetramethylsilane (TMS, assigned 0ppm on δ). In liquids narrow resonance signals are observed with typical widths of 0.1 Hz (Blümich 2005).

The value of using a standard reference is that determining absolute references for nuclei within molecules is not practical. Electromagnetic shielding is a phenomenon influenced by the orbiting electrons producing a local electromagnetic field, in isolation this accounts for nucleus resonance at Larmor frequency. In molecules however, since electrons are shared disproportionally the electromagnetic shielding provided to nuclei with that molecule will be variable, affecting the nuclear magnetic resonance frequency

associated with nature, degree and extent of bonding so provides insight into the extent of unique hydrogen atoms in a given molecule (Macomber 1998). Evaluating the chemical shift of magnetic resonance in relation to a known reference substance is as a ratio of measured difference in frequencies to that reference substance used. In this NMR study, deuterated DMSO (dimethyl sulfoxide-d6) has been used as a reference since commercially available samples are not 100% pure and a residual DMSO-d5 1H NMR signal can be observed at 2.50ppm. (Gottlieb, Kotlyar and Nudelman 1997).

The DSMO was used as an external deuterium lock (not as a solvent), within a capillary tube insert accompanying the sample to prevent contamination. NMR software constantly monitors this known deuterium signal resonance and uses this reference to ensure magnetic field duplicates its known properties.

2.5.5 Coupling constants (J)

NMR spectra usually reflect peaks that occur in groups, due to the interaction of nuclei in close proximity. Individual peaks representing symmetry of molecules, in turn reflect as two or more symmetrical lines representing split patterns (multiplets). The coupling of adjacent particles inherent to magnetic moments (μ) is an important component to providing insight regarding the nature and orientation of molecules studied (Macomber 1998). A hydrogen atom is not able to couple with itself, or with protons that are chemically equivalent e.g. hydrogens in a methyl group, but may couple (up or down) with hydrogen atoms adjacent to the methyl group. This is useful principle that

helps to determine structure of molecules, as it affords perspective to the hydrogen orientation, i.e. the number of Hydrogen atoms involved in unique bonds within a molecule.

Coupling constants guide the interpretation of NMR spectra as to the number of adjacent spin-active nuclei, via the "n + 1" rule, when spin state is ±½, and 'n' = number of protons in different chemical environment, e.g. when there are two neighbouring hydrogen atoms, a triplet peak will result. Again, this is due to the relative interactions within the external magnetic force (B_o), where orientation to this field will impact the measured response and result in the variance in relative chemical shift (δ) (Bakhmutov 2015).

2.5.6 Integration

The area under a peak or group of peaks is directly proportional to the number of spin-active Hydrogen atoms associated to that relative point of magnetic resonance. Although the exact quantity of nuclei can sometimes be difficult to determine, relative comparison is routinely applied to achieve an accurate estimate, so serves as a valuable tool for compound structure determination.

2.5.7 Relaxation times

Spin-active nuclei that experience magnetic resonance at a particular radio frequency (f_1) experience 'spin-flip' and adopt the higher energy orientation to external magnetic force (B_o). Inevitably, once the resonant radio frequency is removed spin-active nuclei will return to their original energy state, dissipating

the activating energy they absorbed, which is measured as inherent to NMR analysis (Reich 2015).

Graphically relaxation times impact the width of peaks displayed on NMR spectra. And homoeopathic dilutions are expected to have relatively narrow and distinct peaks associated with water as a liquid have longer relaxation times (Jacobsen 2007).

T1 Relaxation

Longitudinal / spin-lattice relaxation (T1), refers to restoration of the original energy state within the external magnetic field (B_0), so refers to relaxation of magnetised vector to equilibrium aligned with the 'z-axis' of the static magnetic field (B_0).

The discharge of energy as excited (highly energised) nuclei return to lower equilibrium state is transferred to other surrounding spin-active nuclei and materialised as a general increase of their activity level. For T_1 relaxation to occur there must be magnetic field fluctuations in the x-y direction (Reich 2015).

T2 Relaxation

T2 relaxation is caused by fluctuations in any direction, but is specific as spin-spin / transverse relaxation time, as a measure of the rate of energy exchange in the transverse (x - y) plane. The energy released is redistributed among neighbouring nuclei, which in their turn absorb and transfer this energy.

(Reich 2015). This change is a time constant for a given sample representing the loss of phase-coherence for spins oriented at some angle to the static magnetic field (B_o) (Rinck 2015).

2.5.8 Free induction decay (FID) and Fourier Transform (FT)

Latest NMR spectrometers induce a strong radio frequency pulse that aligns spin-active nuclei causing them to precess in unison (same direction at same time) as Larmor Frequency resonance. Collectively they exert a detectable rotating magnetic field and consequently induce an electrical voltage the Helmholtz RF Coil surrounding the sample. Free Induction Decay (FID) refers to the subsequent deterioration in voltage as nuclei then re-orientate when RF pulse is absent and this diminishing signal contains the collective magnetic resonance frequency date for entire sample (Jacobsen 2007; Simpson and Simpson 2014; Reich 2015).

The computer converts this digitised data using a Fast Fourier Transform to convert it from an FID signal (function of time) into a spectrum in terms of intensity as a function of frequency. NMR “spectrum” yields a peak for all resonant frequencies detected in the sample. Repetitive pulses accumulate and sum FID data which reduces transient ‘noise’ and improves sensitivity of processing. Pulsed Fourier transform process is analogous to playing a chord on the piano and recording the signal from the decaying sound, Fourier transform spectrum would reflect individual notes as ‘frequency peaks’ (Jacobsen 2007).

2.5.9 NMR Summary

^{1}H Nuclear Magnetic Resonance (NMR) is a powerful tool that we can use to derive important information about the nature of a sample being analysed:

- The quantity of Hydrogen atoms with unique magnetic resonance is an indication number of Hydrogen atoms participating in unique bonds, reflected by the number of unique peaks visible on the NMR spectra.
- Chemical shift highlights is a relative reflection of Hydrogen atoms located in unique environments
- According to the 'n + 1' rule the number of neighbouring hydrogen atoms can be determined by identifying the presence of splitting.
- The relative quantity of hydrogen ions as a ppm reading can be derived from integration of those peaks

2.6 NMR Research in Homoeopathy

There is already a significant body of research undertaken at Durban University of Technology (DUT), using NMR to evaluate and compare impact of various aspects on homoeopathic potencies dating back to 1997 (Ross 1997). Just one of these studies, comparing NMR spectra of parallel potencies of Pulsatilla pratensis, prepared according to Hahnemannian and Anthroposophical principles (Erasmus 2004), was performed on a medicinal potency (15% alcohol), while all other studies performed at this institution (Ross 1997; Power 1999; Davies 2001; Cason 2002; Malan 2002; Hofmeyr 2004; Lyell 2004; Botha 2005; Allsopp 2010) were conducted using medicating potencies of 87% or 95%.

Erasmus (2004) found statistically significant differences in both Chemical Shift and Relative Integration values, evaluated using a Varian 500MHz INOVA NMR Spectrometer (Erasmus 2004), which provided an indication that this evaluation performed at 20% alcohol should yield results, samples in this study where measured using the Varian Inova 600 NMR spectrometer.

It is important that the reproducibility of this achievement be evaluated as it is ultimately this alcohol concentration that medicating potency, the prescription a patient receives where the properties inherent to the remedy could have therapeutic significance and where supply chain quality assurance efforts must ultimately be evaluated.

Both Ross (Ross 1997) and Power (Power 1999) suggested a higher frequency may improve sensitivity of the spectrum data obtained. This hypothesis was evaluated using 80MHz, 200MHz and 500MHz nuclear magnetic resonance spectra of sulphur 30CH (Cason 2002), results obtained where shown to be influenced by varied frequency strengths. It is unclear from this study however whether the hardware and software technology associated with these units were identical, so potential impact of technological advancement may have been a factor.

Based on the increasing magnetic field strength associated with these NMR frequencies (in Tesla), a proportional increase in energy differences of spin states associated with increasing frequency should improve spectra resolution achieved (Benesi 2015).

Succussion and trituration are inherent to LM manufacture, while Hahnemannian preparation is central to the purpose of this study. These studies were all able to demonstrate statistically relevant differences well above Avogadro's number, so any variation in application during remedy manufacturing from the various sources reflected on NMR output spectra, as being statistically significant can be assumed to be due to the focus of their investigations. With Natrum Muriaticum, NMR analysis revealed distinct chemical shift and relative integration spectra when the degree of succussion was evaluated, which suggests that potentisation methodology is indeed fundamental to the resulting physicochemical nature inherent to homoeopathic remedies (Lyell 2004)

NMR has been shown to be consistently useful at the high alcohol content of medicating potencies, while only one study (Erasmus 2004) evaluating low alcohol content typical in medicinal potency has been performed at DUT. Current study was undertaken to extend low alcohol content to the physico-chemical properties of LM potency investigate the reproducibility of those results.

In the absence of a scientifically verifiable explanation detailing the action of homoeopathic remedies to substantiate core tenants of 'infinitesimal dose' and 'law of similars', the medicinal benefits for patients realised in practice, will continue to be attributed to the 'placebo effect' and 'psychosomatic' influence, despite many meta-analytical studies indicating otherwise. Often poor quality research is the root cause of failure or poor perception of homoeopathic research, but in high quality studies it has been shown that positive outcomes far outweigh negative (Walach *et al.* 2005).

NMR studies of homoeopathic potencies usually utilise alcohol concentrations typical in medicinal potencies (87% and 95% in DUT studies), which favour NMR study and remedy preservation, but are not representative of posology relevant to patients, where alcohol content is routinely limited to 20% for patient consumption (Kayne 2006). LM prescription requires that the practitioner prepares the patient remedy from the source granule into water, so this study wishes to evaluate the potential to improve quality assurance for that prescription *taken by patient*, using NMR technology in interests of

improved medical care, as it is an ethical imperative that we endeavour to further patient benefit as a primarily motivation in research, as Hahnemann did.

Many other homoeopathic studies using NMR spectra have been undertaken globally, with mixed results, with unable to reproduce previously encouraging results (Aabel, Fossheim and Rise 2001; Anick 2004), citing poor method and contamination (e.g. silica into highly purified water) as being erroneously interpreted as a positive result, which the researcher will attempt to address with the use of borosilicate glassware.

The ability of NMR to reliably detect statistically significant differences in the physico-chemical properties in lactose and water/ethanol base solutions at different potencies was an important development, while establishing a method to leverage this technology has facilitated reproducibility of results. In 1990 a homoeopathic dilution of Sulphur (decimal) challenging Avogadro's number as a constraint observed statistically different relative of H_2O, OH and CH_2 peaks on NMR spectra, using a 300 MHz spectrometer (Weingaertner 1990, 1992). Similar results were demonstrated using a 500MHz Spectrometer to evaluate sulphur at LM potencies in 1997 (Ross 1997). This study initiated considerable further investigation into variables related potentised remedies, including this one using a 600 Mhz spectrometer (Ross 1997; Power 1999; Davies 2001; Cason 2002; Malan 2002; Hofmeyr 2004; Lyell 2004; Botha 2005; Allsopp 2010).

NMR remains a promising technology in researching homoeopathic dilutions as it is capable of detecting structural orientation of molecules (Becker-Witt *et al.* 2003). A central concern related to research and indeed manipulation of homoeopathic dilutions (inclusion of a reference solvent, such as DMSO in this study) is the probability of relatively high contamination of samples (Rao *et al.* 2007). As even a minute amount of foreign material may result in significant scope for error which would impact NMR spectroscopy results; they proposed Raman spectroscopy and Fourier transform infra-red spectroscopy as avenues of investigation for future research.

Studies on homeopathic potencies have an extensive history and results consequently show considerable disparity. One comprehensive comparison of 23 different homeopathic remedies tested across a range of potencies displayed distinctive characteristic not reflected in the placebos (Sachs 1983; Kayne 2006). However, other studies which have been undertaken were not able to replicate the positive outcomes of published NMR studies (Aabel, Fossheim and Rise 2001; Kayne 2006).

A consideration and potential limitation of NMR spectroscopy is that its investigation is limited to detail pertaining to structure of individual atoms in the context of discrete molecular bonding, rather than inclusive view of the impact and interaction of molecular complexes considered at a more global level (Rao *et al.* 2007).

2.7 Other Potential Technologies

Several alternate methodologies for investigating the nature of the physico-chemical properties, inherent to homoeopathic dilutions exist that employ technologies proven to detect and elucidate these properties. These include, but are by no means limited to some promising technologies currently being employed in homoeopathic research of physical properties:

2.7.1 Raman Laser Spectroscopes

- Samples are subjected to a monochromatic light source (i.e. laser) resulting in altered molecular vibrations due to induction of this energy, adaption as scattered light is detected and used in sample identification and quantification. Raman spectrum is a plot of intensity vs frequency of the incident light. (InPhotonics Inc 1999; Rao *et al.* 2007).

2.7.2 Thermoluminescence (Thermally stimulated luminescence)

- Ultra-high dilutions of sodium chloride have been shown to retain the emitted light profile, specific to original salt dissolved after irradiation by X-rays and gamma rays (Rey 2002).

2.7.3 Ultraviolet (UV) Spectroscopes

- A comparative table detailing much of the research into homoeopathic dilutions with UV, visible and/or near infrared light spectroscopy, (where absorption is the measured variable, performed over more than two decades would suggest this technology has much potential (Klein *et al.* 2012).

2.7.4 Perkin Elmer (and Aminco Bowman) spectrofluorometers

- Technology has demonstrated ability to produce precise fluorescence-graph imaging of even the highest dynamization levels. This technique holds considerable potential (Kayne 2006)

2.7.5 Delayed luminescence

- Exploits the phenomenon of extended photon emission after exposure to white light, has demonstrated repeatable and specific characteristics using granules impregnated with homoeopathic potencies. (Lenger, Bajpai and Drexel 2008)

2.7.6 Solvatochromic dyes

- Homeopathic dilutions have been shown to consistently effect colour changes when subjected to solvatochromic dyes. These changes are detected as measured absorbance within the visible spectrum due to these dyes, which change colour according to solvent polarity (Cartwright 2015).

- Biochemical processes involve electron transfer so if homeopathic potencies interact with solvatochromic dyes (as has been demonstrated) it is plausible that homoeopathic potencies similarly interact within the human body to elicit their therapeutic effects.

Physical properties remain a fascinating focus for homoeopathic research, as potential means to discover the underlying mechanisms that would finally explain the anecdotal therapeutic effects witnessed over more than two centuries now. Inevitably some physical variable will then prove useful to

address the current issues in quality assurance and standardisation detailed, so exploration of methodology and potential of available technologies is important as prerequisite. Research continues in a wide array of disciplines that despite controversy and scepticism routinely elicit anomalies not expected in the current understanding of extreme dilution.

There are still many avenues of investigation envisaged in the pursuit of a reliable method for identifying physical properties of homoeopathic preparations. Freezing-point depression, cryo-scanning electron microscopy (cryo-SEM), acoustic loss spectroscopy, ellipsometry, viscosity and studies on surface tension are being explored and may be useful in the future as technology enables study of these physical dimensions and adds to the existing body of work (Rao *et al.* 2007).

Physical research on samples that in theory no longer contain any molecules of the source substance, regularly produce physical characteristics that are not revealed by control samples. This body of research is now extensive and has grown to include a large and expanding range of physical variables being investigated. It would appear that there is much that still needs to be revealed pertaining to the nature and potential of extreme dilution solutions that this foundational work is prerequisite to understanding how remedies are produced and how they elicit therapeutic outcomes within the human body. Laboratory experimentation will need to correlate with clinical application, or even after the post-Hahnemannian construct of the so-called memory of water

(MOW) is definitively explained, the mechanism of therapeutic action would still remain undetermined (Milgrom 2007).

A review of the literature reveals a disturbing trend. A researcher finds an interesting dynamic unexpected in an ultra-diluted substance. In response the homoeopathic community embraces a positive outcome prematurely and disproportionally, while an extremely belligerent community of sceptics and allopathic governing bodies' supress or demonise the research and those researchers associated with it.

The highly controversial example of Jacques Benveniste (Ball 2004), illustrates this typical progression well. He discovered that when solutions containing human antibodies were agitated, they elicited a basophillic response even as ultra-diluted solutions where antibodies were not present. After publishing his results both his funding was withdrawn and he lost his position (Aspinwall 2012). Before his death in 2004 he reflected on his research, and said "Maybe I should have thrown the data away" (Chaplin 2007).

Lack of funding and resources plague this avenue of enquiry (Lewith 2007; Homeopathy Research Institute 2016) so it is perhaps unsurprising that progress correlating the clinical benefits patients experience, has not been reliably reproduced by laboratory experimentation.

2.8 Science of High Dilution

This particular study aligns with the idea that potencies may retain a unique electromagnetic nature despite ultra-dilution (Montagnier *et al.* 2009), which may be detected using NMR Technology and possibly serve as a practical means for evaluation and quality control. Much research has attempted to derive theoretical models that may account for the ability for water to retain an 'intelligence' of crude substance even as dilutions far exceed 'Avogadro's number", the so-called "memory of water" (MOW).

One popular theory proposes that clusters of molecules, which are inherent within a solvent (water) structure, form as hydrogen-bonded water molecules around a solute substance to organise into clathrate structures, to accommodate the introduction of the substance (Anagnostatos *et al.* 1991; Anagnostatos 1994). These clathrate structures are disrupted during succussion due to shear forces, with fragments of these initial clathrates now initiating a similar re-organisation in response to their unique internal conformation, now at a global level within the solvent.

Effectively, this mechanism is proposed to account for retention of the therapeutic effect of the substance solute, even past Avogadro's' constant, when theoretically the source substance is no longer present.

Resch and Gutmann (1991: 231) proposed that water as a solvent has information carrying properties by virtue of its highly ordered "super molecular system". Dissolved contaminants and gases help to shape an ordered lattice

structure. Further, ‘solvation spheres’ develop around hydrophilic solutes, and ‘inner surface’ molecules form around hydrophobic solutes. They postulate that with the aid of agitation such as succussion a substance or solute will modify the water structure, thereby imparting information.

The distinctive orientation and inter-molecular bonding properties of water molecules (H_20) is also the basis of the theory involving ‘structure makers’ (solute) and ‘structure breakers’ (solvent) and resulting ‘void lattices’ (Resch and Gutmann 1987). Organisation within liquids is influenced by phase boundaries between liquid ‘structure makers’ and inner void surface of the gas bubble ‘structure breakers’ which transpose the source remedy imprint throughout the solvent.

A recent paper suggests that potentisation is a ‘quasi-quantum phenomena’ which is inherent in biological systems and homoeopathy (Molski 2011), as the increased concentration of solvent during potentisation is analogous to growth in biological systems and reduces the mean separation between solvent molecules.

CHAPTER 3 - MATERIALS AND METHODS

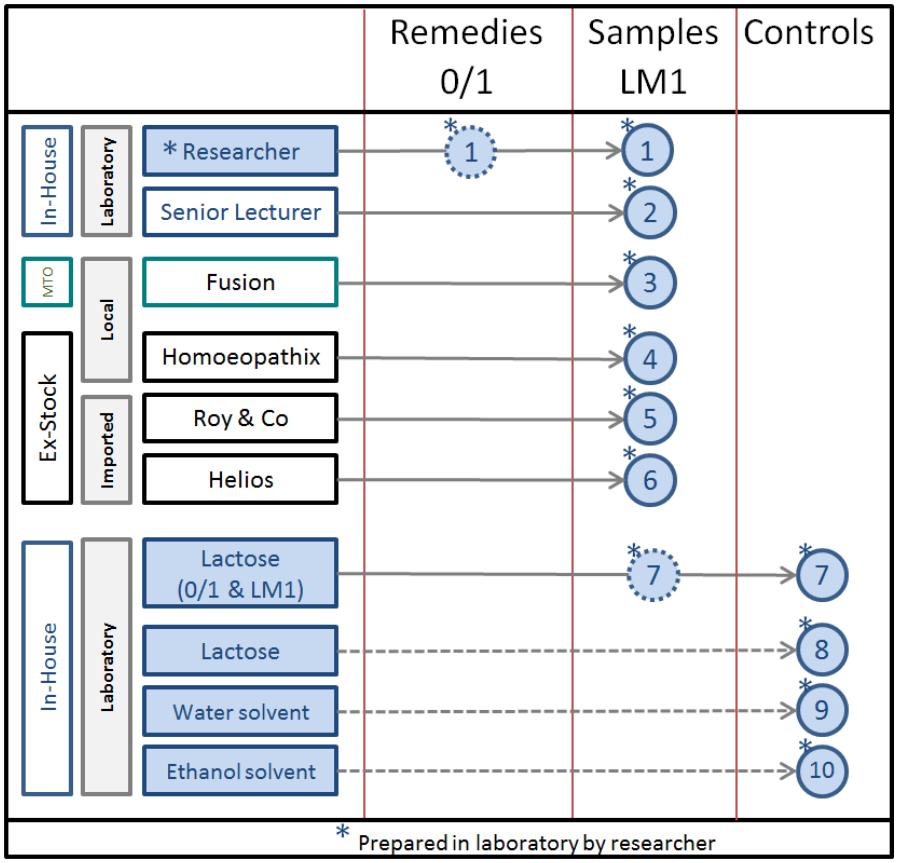

Figure 6: NMR Sample Management

3.1 Introduction

This quantitative study follows a Quasi-experimental design, as the sources of remedies for NMR spectrometry were not randomised, but rather selection was based on availability and convenience. In total, ten samples were sourced and prepared for the Nuclear Magnetic Resonance (NMR) analysis conducted at University of Stellenbosch, using their Varian (Agilent) Unity 600 Mhz NMR Spectrometer. Permission was granted to the researcher in order to prepare NMR analysis experiments personally at the Stellenbosch

University NMR unit laboratory, ahead of performing NMR analysis under supervision of Dr D.J. Brand (Appendix C - NMR Analysis Approval).

In practice, a patient prescription using the LM potency scale involves insertion of a single granule corresponding to that appropriate LM potency into water (Barthel 1991). For this research, we modified this practice to include 20% alcohol (v/v) which would approximate the maximum alcohol content a practitioner would likely include that would serve as a preservative. From a practical standpoint the inclusion was necessary to facilitate the generation of relevant NMR spectra believed to have been successfully implemented in a previous study at medicinal potency of 15% (Erasmus 2004).

Make to Order (MTO) refers to production of a remedy for a specific demand and specification. Three samples where produced which meet this criteria.

- Two In-House remedies produced at Durban University of Technology (DUT) Homoeopathic Laboratory.
    - One remedy produced by Garth Miller (student researcher)
    - One remedy produced by experienced Dr. Izel Botha (Homoeopharmaceutics Senior Lecturer).
- Remedy produced by Fusion Homoeopathics specifically for this research.
- Potentised LM1 (0/1) lactose control sample (no crude substance)

All of these samples utilised the same Lactose Monohydrate and Sodium Chloride inputs (where applicable).

Make to Stock implies production in anticipation of demand, usually to maintain a stock level of a stock keeping unit (SKU) or in anticipation of demand which aligns to a sales forecast. These three LM1 (0/1) samples were supplied to satisfy our order from existing generic stock.

- One sample from Homoeopathix Trading Company (South Africa)
- One sample was from Helios Homoeopathy (UK)
- One sample was from Roy & Company (India)

Given their nature the controls produced could be considered intermediate goods, since they are not 'saleable' in the context of homoeopathic remedies. Lactose, water and alcohol are of course commodities in their own right, but would classify as work in progress (WIP) since they are inputs to the manufacture of these homoeopathic remedies.

3.2 Choice of *Natrum muriaticum* (Nat-m) for this Study

As a crude substance sodium chloride (NaCl), is primarily known for its physiological significance, rather than as a medicinal drug. However, when it is potentised and its energetic potential is liberated, it becomes an important polycrest remedy known as *Natrum muriaticum* (Nat-m) and has profound medicinal value (Banerjee 2006).

Both of the elemental components of salt, sodium and chloride, are essential nutrients vital for life and good health. Sodium (Na^+) is the principal cationic electrolyte within the extracellular fluid (ECF), with its concentration directly proportional to overall water balance. As Na^+ is the major determinant of plasma osmolality, its concentration is carefully controlled by osmoregulation mechanisms for homoeostasis (Koeppen and Stanton 2007). Additionally, sodium is involved in muscle contraction and nerve impulse conduction.

Chloride (Cl^-) is the most abundant anionic electrolyte in the extracellular fluid (Tate and Seeley 2009). Chloride ions are required for production of hydrochloric acid (HCl), which is secreted by the parietal cells in the stomach (Wardlaw, Smith and Collene 2013). HCL serves as an acidic barrier to pathogens and has a digestive function, converting the inactive enzyme pepsinogen into pepsin for protein digestion and also the chemical degradation of food (Caballero, Sadler and Strain 1999).

Originally proved by Hahnemann (Vermeulen 2004) in 1830 (Taylor 2001), *Natrum muriaticum* (Nat-m) is a very important homoeopathic remedy. Nat-m is one of the most routinely prescribed homoeopathic polycrests in practice, since it demonstrates comprehensive mind and body symptoms in provings and clinical experience, consequently Nat-m is represented within thousands of rubrics throughout homoeopathic repertories (Vermeulen 2001; Schroyens and Vint 2007).

Three NMR studies, undertaken at Durban University of Technology evaluated key aspects of homoeopathic remedy potentisation, using *Natrum muriaticum*:

- Succussion and trituration (15CH) (Hofmeyr 2004)
- Extent of succussion on parallel potencies (12CH) (Lyell 2004)
- Hahnemannian vs. radionically prepared potencies (6CH /12CH / 30CH) (Allsopp 2010)

Additionally:

- Using laboratory grade sodium chloride is affordable and of high purity
- Stable nature of this chemical compound allows for relative comparison under variable conditions
- Repeatability of research, if required, is simpler.

Consequently, its choice as the crude substance for this study would easily have been motivated by these factors alone. However, the preparation of LM potencies requires that substances be triturated to 3C prior to preparation of the mother tincture, so strict adherence the Hahnemannian methodology would require this effort. Sodium chloride is however highly soluble in water and this would present a temptation for manufacturers to use the far less labour intensive option of serial dilution. If this indeed proved to be the case this omission could account for any anomalies present on the NMR spectra.

The MTO samples were all produced using laboratory grade Sodium Chloride (NaCl), sourced from Associated Chemical Enterprises, (Batch No. 28584) where their analysis revealed the following results:

Assay	Min 99.5%
Alkalinity (Na_2CO_3)	0.015%
Arsenic (As)	0.00002%
Calcium (Ca)	0.002%
Chlorate and Nitrate (NO_3)	0.003%
Iodide (I)	0.002%
Iron (Fe)	0.0002%
Lead (Pb)	0.00005%
Loss on drying (105ºC)	0.3%
Magnesium (mg)	0.0005%
Sulphate (SO_4)	0.05%
Water – insoluble matter	0.005%

Table 3-1: NaCl Sample Analysis Results

A common criticism of research is that at such high dilution cross-contamination is very difficult to avoid (Anick 2004). No glassware or laboratory equipment in contact with a sample during preparation was re-used to mitigate this potential.

3.3 Sourcing of Remedies

3.3.1 MTO - DUT LM1 (0/1) Nat-mur samples

Two samples of *Natrum muriaticum* LM1 (0/1) were produced in-house at Durban University of Technology in the Homoeopathic Department Laboratory in accordance to German Homoeopathic Pharmacopoeia (GHP 17) (*German Homoeopathic Pharmacopoeia* 1985). This methodology was aligned to be in accordance with the methodology prescribed by Hahnemann in his 6th edition of the *Organon* (Aphorism 270) for LM remedy as discussed in appendix: Preparation of Natrum muriaticum LM (0/1).

Hahnemann used the accepted measurement scale prevalent to that period (Apothecary's measures in 1802). After performing a literature review we established that Hahnemann's reference to a Troy grain was equivalent to 0.0648 grams and furthermore equivalent to 1 drop (ggt).

Subsequently Dr Botha further researched this aspect for her paper presented at the LIGA 2014 Conference in Paris (Botha 2014). Her findings were that 0.053g would be a more accurate approximation representing Hahnemann's reference measurements, under the French Apothecary system. If this is indeed true, despite attempting to rigorously follow Hahnemann's method exactly, the methodology employed here fails to do that (Barthel 1991; Hahnemann and O'Reilly 2001). She further found that Hahnemann's reference to "drops" was most likely equivalent to 1 grain in the English Apothecary system, used during the manufacture of these samples. Given

the difficulty establishing the original quantities and volumes used by Hahnemann it is little wonder standard practice has over time deviated from the original methodology Hahnemann prescribed.

During manufacturing, laboratory conditions were carefully controlled to prevent the introduction of unanticipated variables that would influence the accuracy of the manufacturing process and unduly influence results. Both DUT MTO remedies were produced at the same time, under the same conditions, and with same inputs and proportions at each stage.

- With same inputs and environment, would experience and technique yield a discernible difference in the NMR spectra?
- Hahnemann considered technique to be especially important during the trituration process (Barthel 1991; Hahnemann and O'Reilly 2001).

As the Homoeopharmaceutics Senior Lecturer, Dr Botha was able to ensure that best practice in laboratory standards and procedures were applied at each stage during remedy production.

MTO samples were all manufactured using:

- Laboratory grade Lactose Monohydrate ($C_{12}H_{22}O_{11}.H_2O$), sourced from Merck (Pty) Ltd, (Batch No 1039504), in order to ensure quality complied with requirements outlined in the European Pharmacopoeia 4.
- Liquid potencies were all produced using the same batch of purified distilled water (AqDist), prepared by DUT Laboratory.

- The mass of three separate lots of 100 granules were measures, then the average of those three measurements were used to establish a suitable average mass for “100 grains” referred to in Aphorism 272 (Hahnemann and O'Reilly 2001). This figure of 0.8585 grams was then used to standardise the manufacturing process, as specific to this batch and granule size distribution.
- Laboratory grade Sodium Chloride (NaCl), sourced from Associated Chemical Enterprises (Batch No. 28584), which complies with requirements outlined in the European Pharmacopoeia 4 (See Table 3-1: NaCl Sample Analysis Results).
- Anhydrous Alcohol 99.9%, sourced from Illovo Sugar Limited, (Batch No. 52/12/67) was used to produce alcohol concentrations utilised in manufacture of all MTO samples diluted using purified distilled water (AqDist). Again, this alcohol, which would also be utilised to produce all the remedy samples for NMR analysis, met requirements outlined in the European Pharmacopoeia 4.

For a detailed account of this methodology, please refer to Appendix B, “Preparation of *Natrum muriaticum* LM (0/1)”.

3.3.2 MTO - DUT LM1 (0/1) Lactose Sample

As a reference for comparison of NMR spectra, a LM1 (0/1) Lactose sample was included as a control. This sample was manufactured to exactly the same methodology outlined in Appendix B, “Preparation of Natrum muriaticum

LM (0/1)"; except that 0.065 grams of Lactose also served as the initial crude substance (i.e. replaced sodium chloride (NaCl) as the crude substance).

Since manufacturing inputs, technique and laboratory environment are constant, we propose that the comparison variable in NMR analysis is specific to the crude substance alone, since dilution (5.5CH) falls just short of the limitations imposed by Avogadro's number 6CH) (Morgan and Helios Pharmacy 2014).

3.3.3 MTO - Fusion Homoeopathics LM1 (0/1) Nat-mur samples

This is a make to order (MTO) requirement, in order to evaluate the impact of environment and manufacturing practices for a given sample on NMR spectra.

To reduce the number of variables, the following raw material used in the production of the DUT sample was provided to Fusion Homoeopathics:

- Laboratory grade Sodium Chloride (NaCl), sourced from Associated Chemical Enterprises, (Batch No. 28584)

In comparison to DUT, some notable differences in the methodology are evident, for details refer to documents in MTO - Fusion Homoeopathics cc - Trituration's, and MTO - Fusion Homoeopathics cc - LM Potencies:

- Trituration (Manufacturing Method HAB6 cited)
- Sample quantities were tripled.
- Ambient temperature was noted at 14ºC (DUT ambient temp 22ºC).

- LM0 (Manufacturing Method HAB17 cited)
- 0.6g of 3C trituration was added to 200ml of ethanol at 15% (m/m) during the manufacture of LM0 tincture.
- 1 drop of the above solution to 2.5ml of 86% ethanol (m/m) which is succussed 100 times.
- The solution is then used to “consistently impregnate” 100g of size 10 granules, which are allowed to air dry.
- Water in used in the manufacture has been ozonised.

This company specialises in producing remedies on demand and to specification, where highly experienced and qualified practitioners take personal responsible for the manufacture of their products.

3.3.4 MTS – Homoeopathix cc. LM1 (0/1) Nat-mur

Order was delivered using the services of a courier.

Their manufacturing document states that it is based on 6th Edition of the *Organon, please* refer to MTS – Homoeopathix Manufacturing Document.

Here 0.0060g of 3C triturate is dissolved in 15ml of AqDist before 15ml of 96% ethanol is added (1:1 ratio).

- 1 “squirt” (approximately 15 drops) of 96% ethanol equating to 3.6ml is added to 1 “drop” of LM0
- Succussed 100 times.

- Labelled as LM1

A single “size 0 sucrose granule” is impregnated using 1 drop of the relevant preceding liquid potency, LM1.

3.3.5 MTS – Helios (UK) LM1 (0/1) Nat-mur

Several previous attempts to import sample from companies abroad were thwarted by customs or delivery issues. This remedy was procured online and couriered as a personal favour to circumvent irradiation and customs restrictions.

LM0

- 1 drop of 3C is added to about 17ml of 96% ethanol
- Gently swirled to mix and labelled as LM0 on the cork

LM1

- One drop of LM0 is added to 3.4ml of 96% ethanol
- Succussed 100 times.

LM1

- 2 gram vial is used to measure and decant ‘poppy seed’ lactose granules into filter paper (folded into cone and suspended)

- The ‘poppy seed’ lactose granules are impregnated using 10-12 drops of LM1 medicating potency.
- Filter paper is folded flat and remedy left to dry
- Utilise a culture tube for succussion

3.3.6 MTS – Roy & Co. (India) LM1 (0/1) Nat-mur

The Roy & Co. sample was provided by the DUT dispensary as one of their preferred imported remedy suppliers, for this evaluation. 2 granules were donated where the following details were on the label.

- (Batch DL No. HL-805-M / Manufactured October 2010).

The company was subsequently contacted for the relevant manufacturing documents however they were unable to comply, since they do not produce the remedies themselves and were unable to trace back to the original manufacturer. Roy & Co. operations are limited to repackaging and distributing remedies.

Consequently, the conditions under which this sample was produced remain unknown. This highlights the conditions practitioners routinely encounter when sourcing remedies, even from well-established suppliers who may serve as agents to manufacturers. After consideration, it was decided to include this sample in this study in order to evaluate the possible impact of sourcing a remedy which is considered good quality, but where manufacturing standard practice is assumed but unknown.

3.3.7 Controls

The controls were prepared immediately ahead of NMR Analysis in the Stellenbosch University Chemistry Department Laboratory. The water utilised was sourced from the DUT Homoeopathic Laboratory and transported in borosilicate glass jars.

a) Alcohol Sample

A bottle was filled with Anhydrous Alcohol 99.9%, sourced from Illovo Sugar Limited, (Batch No. 52/12/67) as used to produce alcohol concentrations and as a control sample for analysis. The basis for its inclusion was to ensure no significant contamination existed. The impact of such contamination is that it would affect almost all the samples, since it was used in the product of final medicinal potencies, evaluated during NMR analysis. The following equation was used to calculate the alcohol concentration conversions from this source:

$$A_{Vol1} \times A_{\%} = B_{Vol2} \times B_{\%}$$

A_{Vol1} = desired volume $A_{\%}$ = desired % ethanol

B_{Vol2} = volume on hand $B_{\%}$ = on hand % ethanol

Equation 1: Ethanol Concentration Equation (Banerjee 2006; Kayne 2006)

b) Water Sample

Purified Aq Dist from DUT was transported to Stellenbosch for use in sample preparation and as a control sample for analysis. The basis for its inclusion

was to ensure no significant contamination existed and that same H_2O batch was used throughout. The impact of such contamination is that it would affect almost all the samples, since it was used in the production of final medicinal potencies evaluated during NMR analysis.

Additionally, this water source (same batch from same container) was used in the production of both the DUT – MTO remedies and the DUT – Lactose LM1 Control, so utilising the same water eliminated water as a variable in these instances.

c) Lactose Sample

0.065g of Lactose Monohydrate, the same batch used in the trituration of the DUT samples, was dissolved within 110ml purified Aq Dist per the sample details above.

3.3.8 Transport to Cape Town

The various sources of LM1 0/1 *Natrum muriaticum* remedy sample granules were transferred into appropriately labelled 5ml screw top bottles.

LM1 0/1 lactose and lactose sample granules were similarly transferred into appropriately labelled 5ml screw top bottles.

Ten unused 250ml Borosilicate bottles and lids were meticulously cleaned using detergent, and then thoroughly rinsed using copious amounts of water. Each bottle was then cleaned with 95% alcohol and flamed, process was repeated twice. Bottles and lids were autoclaved twice, allowed to cool before nine were slowly filled completely with distilled water; the tenth bottle was filled

with 95% alcohol, before being wrapped in tissue paper and boxed. All liquids were filled completely as suggested by Barthel (1991) to avoid potential succussion should an air space of consequence remain (Barthel 1991).

All samples were carefully packed into a 'carry-on' bag wrapped in tissue along with copious sachets of silica gel, prior to the approximately two hour flight from King Shaka International to Cape Town International airport. None of the samples were exposed to x-ray examination, instead superficial manual inspection was deemed acceptable by airport security upon request. Care was taken to protect samples from environmental noise, vibration, temperature, light and electromagnetic influence as far as this was practical.

These precautions exceed those considered acceptable practice within the homoeopathic community. Both local and imported remedies are routinely despatched to practitioners via courier or postal services, with only minimal packaging required to protect contents from breakage.

This is a logical approach as homoeopathic remedies are commonly carried by patients and remain effective therapeutically. Additionally, homoeopathic remedies are routinely manufactured, or produced by practitioners, without exhaustive measures to mitigate environmental conditions.

The extraordinary measures undertaken to protect these samples were aimed at reducing the impact of variables as much as was practical, as was done in previous NMR research (Power 1999; Lyell 2004; Botha 2005; Marsh-Brown

2016), to reduce possible impact of transportation on NMR sample analysis undertaken as much as was practical.

3.4 Preparation of samples for NMR Analysis

In total ten samples were prepared for analysis. Each sample was to be analysed five times and consequently, fifty unique samples were prepared in NMR sample tubes.

3.4.1 Potentised Samples

The six Natrium muriaticum LM1 samples were produced from the Natrium muriaticum LM0 (0/1) samples, according to LM1 sample for analysis. Similarly the Lactose LM1 control followed the same procedure.

The 20% alcohol concentration was produced in accordance to the following equation from Alcohol Sample in 3.4.2 below.

$$A_{\text{Vol1}} \times \text{A}_{\%} = \text{B}_{\text{Vol2}} \times \text{B}_{\%}$$

$$700ml \times 20_{\%} = \text{B}_{\text{Vol2}} \times 99.9_{\%}$$

$$140.14ml = \text{B}_{\text{Vol2}} \text{ Aq Dist}$$

Equation 2: Producing 20% ethanol from 99.9% Ethanol Sample

Sample volume of 118.294ml at 20% alcohol was considered closer adherence to Hahnemann's exact method outlined in Aphorism 270 of the 6th Edition *Organon* (Hahnemann and O'Reilly 2001) which is why 110ml was not

used as was the case with other study performed at this alcohol concentration range (Erasmus 2004), which was performed using 15% alcohol.

This emulates standard practice within homoeopathic community where LM1 Medicinal potencies are produced at this alcohol concentration level for patient consumption.

3.4.2 Alcohol Sample

A 30 ml amber glass bottle was filled with Anhydrous Alcohol 99.9%, sourced from Illovo Sugar Limited, (Batch No. 52/12/67)

3.4.3 Water Sample

Purified Aq Dist from DUT was transported to Stellenbosch for use in sample preparation. The quality and purity of this water source was not analysed. Any impact water quality may have was mitigated by using this water source (collected at the same time), for preparation of all samples ahead of NMR analysis, so the impact would be standardised throughout the samples analysed.

There was no requirement for a high purity or laboratory grade water source envisioned or detailed in Hahnemann's methodology, as his standard is implicit to this study it was not deemed necessary.

Certainly, there is no expectation that the demanding ISO Laboratory Water Quality Standards, outlined in Figure 7 – ISO 3996 Standards for Laboratory Water, were represented or required, since provings were conducted long before these standards were established or even practically feasible.

Parameter	Grade 1	Grade 2	Grade 3
Conductivity µS/cm (temp corrected)	< 0.1	< 1	< 5
pH at 25°C	N/A	N/A	5.0 - 7.0
Oxidizable matter Oxygen (02) content mg/L	N/A	< 0.08	< 0.4
Absorbance at 254 nm and 1 cm optical path length, absorbance units	< 0.001	< 0.01	N/A
Residue after evaporation on heating at 110°C mg/kg	N/A	< 1	< 2
Silica (Si02) mg/L	< 0.01	< 0.02	< N/A

Figure 7: ISO 3996 Standards for Laboratory Water (Puretec Industrial Water 2011)

3.4.4 Lactose Sample

0.065g of Lactose Monohydrate, the same batch used in the trituration of the DUT samples, was dissolved within 110ml purified Aq Dist per the sample details above.

3.4.5 Deuterated DMSO

The Deuterated DMSO insert was prepared and added to a separate capillary tube. This capillary tube was inserted into the NMR sample tube along with each sample being analysed, which ensured that sample was not compromised by contamination. Its presence served as a reference substance against which the NMR control logic could use as a calibration reference during readings. As can be seen in the spectra in Figure 8 below the DMSO has a peak at 2.50 ppm.

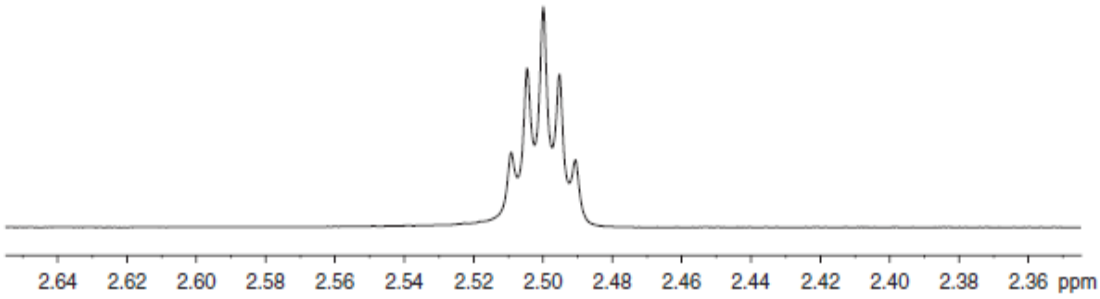

Figure 8: Residual solvent signal in DMSO (Richards and Hollerton 2010)

According to Richards and Hollerton (2010), the biggest challenge with DMSO is its affinity for water, making it almost impossible to keep dry and therefore the use of sealed 0.75 ampoules is recommended. The D6-DMSO as a result will invariably have an enlarged water peak. Another characteristic is its ability to absorb through skin, so unless extreme caution is taken when handling DMSO solutions, your sample can be hazardously absorbed (Richards and Hollerton 2010). Consequently this insert was prepared by Dr DJ Brandt in order to comply with safety and handling procedures pertaining to his laboratory.

3.4.6 Water Quality

It is acknowledged that even high purity laboratory water source is a potential source of contamination, when conduction ultra-dilution analysis.

The same water was used for preparation of DUT MTO and Lactose LM1 samples. Additionally, this same batch of water was transported to Cape Town in order to prepare all liquid samples from LM1 granules for analysis.

3.4.7 Randomisation

After the NMR were analysed in alphabetical sequence, the samples were reconciled to their assigned alphabetical identity as outlined in table 3-2 below.

Assigned	Sample Description
A	MTO - DUT Garth Miller
B	MTS - Roy & Co.(India)
C	MTS - Helios (UK)
D	MTO - Izel Botha
E	CONTROL - Lactose LM1
F	MTO - Fusion Homoeopathics (South Africa)
G	MTS - Homoeopathix Trading Co. (SA)
H	CONTROL – Lactose
I	CONTROL - 95% ROH
J	CONTROL – H_2O

Table 3-2: Randomisation Table.

3.5 NMR Analysis

3.5.1 Final NMR Tube Preparation

The researcher prepared the final fifty samples for NMR analysis while at the Stellenbosch University NMR unit laboratory, ahead of performing NMR analysis under supervision of Dr D.J.Brand.

- A new micro pipette tip was used for each of the ten samples which were blinded and labelled A to J.

- Each sample was used to transfer a volume of 750µl to five new NMR sample tubes.

3.5.2 NMR Processing of Samples

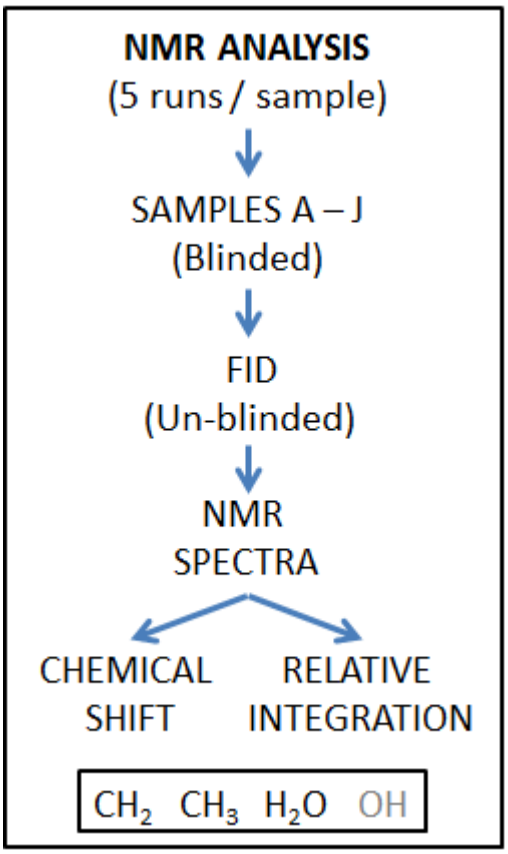

Figure 9: NMR Method Overview

An Agilent ^{Unity}*Inova* 600 NMR Spectrometer ® (Stellenbosch - Central Analytical Facilities (CAF) 2015) was used to analyse the prepared samples. The following parameters prevailed during the data acquisition:

Parameters	
Parameter	**Value**
Origin	Varian
Spectrometer	inova
Solvent	dmso
Temperature	25.0 (ºC)
Pulse Sequence	s2pul
Probe	idpfg
Number of Scans	16
Receiver Gain	40
Relaxation Delay	2.5
Pulse Width	7.1
Acquisition Time	2.7308
Spectrometer Frequency	599.99
Spectral Width	5999.7

Table 3-3: NMR Parameters

Dr DJ Brand initiated this NMR evaluation by careful review and analysis of similar research undertaken by DUT, which utilised the Varian 500 NMR spectrometer in the Chemistry Department of the University of KwaZulu Natal. Calibration and parameters were aligned to suit the nature of homoeopathic dilutions, to ensure results attained would build on the body of work described in Chapter 2 - NMR Research in Homoeopathy.

Each sample was subjected to the same parameters and once the software sequences were configured, manual repetition related to supporting data acquisition followed a routine sequence:

- The DSMO insert was introduced to the NMR sample tube containing the randomly blinded samples (labelled A-J).
- The sample was closely inspected to ensure the DMSO insert was properly immersed and absence of air bubbles.
- In practice, the standard lactose sample and the ethanol sample were clearly distinguishable from the other samples, which effectively circumvented the double-blinding protocol within the research methodology for these samples.
- This is an inherent issue, initially avoided by having samples processed completely independently; however became a prerequisite requirement that researcher was present as an active participant during testing, during the Research Proposal Process.
- NMR tube was inserted through the supporting plug and the tube aligned carefully using the customised template for this purpose
- This ensured the ideal alignment of the sample within the magnetic field.
- The NMR unit was placed in the sample ejection mode, implying a positive displacement of air available to suspend the sample at the superior opening of the feed tube.
- Once the sample was stable, the software initiates the loading sequence and the sample is lowered and induced to revolve within the sensors at the terminal end of the probe positioned from the bottom of the feed tube.

- Before each sample was taken, the magnet was ‘shimmed’ to ensure a homogenous field (B_0) was maintained, in relation to the DMSO reference
- The complexity of this equipment is concealed within the state-of-the-art engineering technological capabilities of the supporting hardware and software.
- This routine was repeated for all fifty samples.

3.6 FID Data

The standard observable NMR signal generated by precessing non-equilibrium nuclear spin nuclei about the constant magnetic field takes form as a discrete digital time domain sampling sequence known as a free induction decay (FID), detailed in Chapter 2 - Free induction decay (FID) and Fourier Transform (FT). The most common approach to convert this ‘time domain signal’ into a frequency domain NMR spectrum is the application of a Fourier transformation (Simpson and Simpson 2014).

3.6.1 NMR Spectra

This FID data was recorded onto a USB Flash Drive, with an additional backup sent to an online file storage service. In order to process and interface with the FID file format, MestReNova Version 10.0.02 was employed, which was the most current version of this multivendor, multiplatform software

used to visualize, process, analyse and report NMR FID data (Mestrelab Research 2014).

For each sample, a graphical representation (NMR Spectra) was derived in order to enable comparison and statistical analysis implicit to this research. Reliability of NMR interpretation that relies on the determination of the relative integral intensities and chemical shift, is dependent on the accuracy and quality of NMR spectra that are used in their determination, which requires processing that ensures accurate phasing and baseline correction after Fourier transformation (Bakhmutov 2015).

For each sample, the graph was processed as follows:

- Phase correction was applied using global Whitening algorithm.
- Baseline correction and smoothing, utilise the Whittaker Smoother.
- Peaks were normalised to referent solvent DSMO
    - Individual peak at 2.500 ppm was universally referenced to 0.2, as this ensured all graphical peaks were comparable across samples.
- Integration derivative algorithm applied to peaks was based on sum, using auto-detection.
- For each graph, the reference peaks were labelled
    - H_2O (HDO)
    - CH_3
    - DSMO
    - CH_3

Note:

OH peak was not demonstrated at only 20% concentration.

The fifty detailed graphs (see NMR Spectra) form the basis for detailed analysis used to derive the comprehensive set of raw data input for statistical analysis. Chemical shift and relative integration of the H_20, CH_2 and CH_3 peaks are the focus of this research, which aligns with the methodology to the body of work already shown to be effective at DUT. The absence of an OH peak is noted and ascribed to the low concentration of ethanol (20%) of this study.

3.6.2 Chemical shift

- Chemical Shift values were captured into a Microsoft Excel 2010 spreadsheet to facilitate processing (see Summary of data obtained from NMR spectroscopy).
- The multiple peaks associated with CH_2 and CH_3 were averaged to assign a single value to represent there profile.

3.6.3 Relative Integration

- Relative Integration values were captured into a Microsoft Excel 2010 spreadsheet to facilitate processing (Summary of data obtained from NMR spectroscopy).
- This sum of the areas below each peak was divided by the total area under the collective peak values

3.7 Statistical Analysis

The values for Chemical Shift and Relative Integration where used as input for statistical analysis in GNU PSPP version 085, a free replacement for the proprietary program SPSS (Free Software Foundation 2015). The data was analysed using descriptive statistics and subsequently non-parametric tests (distribution-free tests) (Stewart 2002), necessitated by the small sample size (n) and since distribution shown not to be normally distributed. This methodology aligns with previous studies undertaken at DUT (Ross 1997; Power 1999; Davies 2001; Cason 2002; Malan 2002; Hofmeyr 2004; Lyell 2004; Botha 2005; Allsopp 2010).

3.7.1 Kruskal-Wallis

The Kruskal-Wallis Test (Kruskal-Wallis H Test) is a non-parametric alternative to a one-way ANOVA for assessment of between-groups analysis of variance. It is similar in nature to the Mann-Whitney test, but it allows comparison of more than just two groups and is well suited to compare scores based on continuous variable found in three or more groups (Pallant 2001; Field 2013). Values are converted to ranks and the mean rank for each group is then compared. Given that this is a 'between-groups' analysis, different data points people must be in each of the different groups.

Key outputs are:

- Chi-Square value
- Degrees of freedom (df)

- Significance level (presented as Asymp. Sig.).

If this significance level is a value less than .05 one can conclude that there is a statistically significant difference within the continuous variable across the three groups (Pallant 2013). Kruskal-Wallis test was selected as the distributions within the ten groups were not normal, and the sample sizes were small. While able to determine that overall if there is statistically relevant differences in the data, it does not identify the source or distribution of this difference (Field 2013).

Kruskal-Wallis uses ranked data where the data is collated and ordered from smallest to largest value (amorphous, in the source group is not retained). This order is then ranked numerically starting with '1' for lowest value through to highest ('n'). The data is re-assigned to original datasets along with new assignment, but then rank assigned rather than the original value is used to calculate sum (R_i in Equation 3) and average of each sample set.

$$H = \frac{12}{N(N+1)} \sum_{i=1}^{k} \frac{R_i^2}{n_i} - 3(N+1)$$

Equation 3: Test Statistic H in Kruskal-Wallis (Field 2013)

In Equation 3,

- 'N' represents the total sample size (in this case 50).
- n_i is the sample size in particular group (in this case 5).

- k groups present in the data (in this case 10).
- R_i represents the sum of the ranks for groups identified by 'i'.

The significance (α) has been set at 0.05

- H_0: There is no statistically relevant difference between the LM1 remedy samples.
    - Accept H_0: if p value is ≥ α
- H_1: There is a statistically relevant difference between the LM1 remedy samples.
    - Accept H_1: if p value ≤ α

3.7.2 Mann-Whitney Test.

The results of Kruskal-Wallis analysis demonstrated a statistically significant difference within the data. Consequently the non-parametric individual sample pairs were next evaluated using the Mann-Whitney test (comparing medians), which can be considered as the non-parametric equivalent of the independent t-test (which compares means) of the two groups (Pallant 2013).

Method

- As with Kruskal-Wallis, values from the two sample groups assessed are sorted from lowest to highest value, the groups of origin are disregarded.

- Lowest value is assigned a rank of 1, the next highest value a rank of 2, with this process repeated incrementally until all values are ranked.
- When two (or more) values have equal value, all those values share one ranking
- This value is calculated as a mean of their accumulated rankings, based on sequential assignment.
- The sample data is again assigned to their source groups, together with the rankings derived
- All the scores with their assigned rankings are then regrouped into the sample groups they originated from
- The sum of the rankings in group R_1 and group R_2 are calculated.
- The Mann-Whitney test is evaluated according to the following formulae to calculate U (Equation 4):

$$U = n_1 n_2 + \frac{n_1(n_1 + 1)}{2} - R_1$$

Equation 4: Test Statistic U in Mann-Whitney (Field 2013)

In Equation 4:

- n_1 n_2 are sample sizes of sample groups 1 and 2 being compared
- R_1 is the sum of ranks for group 1.

The key output values after analysis is the Z value

- Significance level as Asymp. Sig (2-tailed)
- The smaller of U value of the two groups (as the p value) was used and compared to significance value (α).

The significance (α) has been set at 0.05

- H_0: There is no statistically relevant difference between the LM1 remedy samples.
    - Accept H_0: if p value is ≥ α
- H_1: There is a statistically relevant difference between the LM1 remedy samples.
    - Accept H_1: if p value ≤ α

CHAPTER 4 - RESULTS

4.1 Criteria Governing the Admissibility of Data

Every practical precaution was implemented to avoid contamination and environmental conditions which could potentially impact on the nature or electromagnetic properties of the samples for NMR analysis. Half of the *Natrum muriaticum* remedies were procured however, so were subject to environmental influences typically associated with storage and handling, wherever possible these were mitigated using couriers. This is representative of conditions prevailing when practitioners place orders for deliver, which is a routine activity.

For all the make-to-order samples, strict laboratory best practices were applied.

From the spectra produced by the NMR analyses, raw data was obtained and used to determine the chemical shift (δ) values and to calculate the relative integration values.

Comparisons of the chemical shift (δ) and relative integration values for the H_2O, CH_2 and CH_3 peaks were performed between all the experimental and control groups. The comparisons were performed by means of statistical analysis as set out in section 3.7 of chapter three.

The analysis of the solvent control samples showed no contaminants or significant differences between the five samples run for each. No artefacts that could be assigned to the controls were identified on the *Natrum muriaticum* LM1 NMR spectra.

Only four readings were registered for the Lactose LM1 - Control sample, the third of the five readings was not successfully recorded. It remains unclear as to how this data point attained a null value, but this anomaly was only evident once the FID files were processed graphically. Consequently it was decided that only these four values would comprise the statistical analysis for this sample.

4.2 Chemical Shift (δ) Values

4.2.1 Descriptives of chemical shift (δ) values

Valid cases = 36; cases with missing value(s) = 2.

Variable	*N*	*Mean*	*Std. Dev*	*Variance*	*Kurtosis*	*S.E. Kurt.*	*Skewness*	*S.E. Skew*	*Range*	*Min.*	*Max.*
H_2O	34	4.42	.01	.00	-.95	.79	**.16**	.40	.03	4.41	4.44
CH_2	34	3.16	.00	.00	-.73	.79	**.26**	.40	.01	3.16	3.17
CH_3	34	.69	.00	.00	-.57	.79	**.33**	.40	.01	.69	.70

Table 4-1: Descriptives Statistics of Chemical Shift

Data is skewed, so shown to be 'not normally distributed', so parametric tests could not be performed, as only non-parametric analyses are applicable. The chosen non-parametric tests were Kruskal-Wallis and Mann-Whitney.

4.2.2 Kruskal-Wallis Test: Comparison of chemical shift (δ) values

	H_2O	CH_2	CH_3
Significance (α)	**0.000**	**0.000**	**0.000**

Table 4-2: Kruskal-Wallis Test: Comparison Of Chemical Shift (δ) Values

Significance values sourced Chemical Shift Analysis Of Variance (ANOVA).

The comparison of the chemical shift values shows a significant difference between all 5 samples in all three peaks, though does not describe the nature of this difference (where these differences manifest).

The hypotheses are thus both accepted.

- OH Peak was not represented as expected based on a similar NMR study at 15% (Erasmus 2004).
    - At the 20% ethanol concentration utilised in this study, there is insufficient ethanol present to manifest as an OH peak as seen in studies conducted at medicating potencies of greater than 85% ethanol.
    - It was hoped that the higher specification of the Agilent ^{Unity}*Inova* 600 NMR Spectrometer® (Comparable study used the Varian 500MHz INOVA spectrometer (Erasmus 2004)) and a slightly higher alcohol concentration of 20% would contribute to include this peak.

4.2.3 Mann-Whitney Test: Comparison of chemical shift (δ) values

Data reflected in the Independent T-test (not assumed since data is not normally distributed), values extracted from Sig. (2-tailed).

a) Control Lactose LM1 and MTO Garth

	H_2O	CH_2	CH_3
Significance (α)	.002	.072	.004

Table 4-3: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTO Garth

The comparison of the chemical shift values of the Control Lactose LM1 and MTO Garth groups show a significant difference for the H_2O and CH_3 peaks.

The hypotheses are thus accepted.

b) Control Lactose LM1 and MTS Roy & Co.

	H_2O	CH_2	CH_3
Significance (α)	.000	.007	.004

Table 4-4: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTS Roy & Co.

Comparing the Control Lactose LM1 and MTS Roy & Co. groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

c) Control Lactose LM1 and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.002	.000	.000

Table 4-5: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTS Helios.

Comparing the Control Lactose LM1 and MTS Helios groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

d) Control Lactose LM1 and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.000	.016	.000

Table 4-6: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTO Izel.

Comparing the Control Lactose LM1 and MTO Izel groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

e) Control Lactose LM1 and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.012	.002	.000

Table 4-7: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTO Fusion Homoeopathics

Comparing the Control Lactose LM1 and MTO Fusion Homoeopathics groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

f) Control Lactose LM1 and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.188	.003	.000

Table 4-8: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTS Homoeopathix.

The comparison of the chemical shift values of the Control Lactose LM1 and MTS Homoeopathix groups show a significant difference for the CH_2 and CH_3 peaks.

The hypotheses are thus accepted.

The H_2O values were not shown to be significantly different, so hypotheses for this peak is thus rejected

g) MTO Garth and MTS Roy & Co.

	H_2O	CH_2	CH_3
Significance (α)	.064	.031	.421

Table 4-9: Mann-Whitney Test: Chemical Shift (δ) Values for Control Lactose LM1 and MTS Roy & Co..

The comparison of the chemical shift values of the MTO Garth and MTS Roy & Co., groups show a significant difference for CH_2. The

hypotheses are thus accepted. The CH_3 and H_2O peaks are not significantly different; hence hypotheses for these values are thus rejected.

h) MTO Garth and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.002	.000	.000

Table 4-10: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Garth and MTS Helios.

Comparing the MTO Garth and MTS Helios groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

i) MTO Garth and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.053	.374	.456

Table 4-11: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Garth and MTO Izel.

Comparing the MTO Garth and MTO Izel groups, the chemical shift values show no significant difference between all the peaks.

The hypotheses are therefore rejected.

j) MTO Garth and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-12: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Garth and MTO Fusion Homoeopathics.

Comparing the MTO Garth and MTO Fusion Homoeopathics groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

k) MTO Garth and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.001	.000	.000

Table 4-13: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Garth and MTS Homoeopathix.

Comparing the MTO Garth and MTS Homoeopathix groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

l) MTS Roy & Co. and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.088	.000	.000

Table 4-14: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Roy & Co. and MTS Helios.

Comparing the MTS Roy & Co. and MTS Helios groups, the chemical shift values show a significant difference between CH_2 and CH_3 peaks. The hypotheses are thus accepted. H_20 does not show a significant difference; hence hypothesis for this value is rejected.

m) MTS Roy & Co. and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.787	.062	.822

Table 4-15: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Roy & Co. and MTO Izel.

Comparing the MTS Roy & Co. and MTO Izel groups, the chemical shift values do not show a significant difference between all the peaks.

The hypotheses are thus rejected.

n) MTS Roy & Co. and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-16: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Roy & Co. and MTO Fusion Homoeopathics.

Comparing the MTS Roy & Co. and MTO Fusion Homoeopathics groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

o) MTS Roy & Co. and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.001	.000	.000

Table 4-17: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Roy & Co. and MTS Homoeopathix.

Comparing the MTS Roy & Co. and MTS Homoeopathix groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

p) MTS Helios and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.074	.000	.000

Table 4-18: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Helios and MTO Izel.

Comparing the MTS Helios and MTO Izel groups, the chemical shift values show a significant difference between CH_2 and CH_3 the peaks. The hypotheses are thus accepted. There is no significant difference for H_2O peaks; hence hypothesis for this value is rejected.

q) MTS Helios and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-19: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Helios and MTO Fusion Homoeopathics.

Comparing the MTS Helios and MTO Fusion Homoeopathics groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

r) MTS Helios and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-20: Mann-Whitney Test: Chemical Shift (δ) Values for MTS Helios and MTS Homoeopathix.

Comparing the MTS Helios and MTS Homoeopathix groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

s) MTO Izel and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-21: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Izel and MTO Fusion Homoeopathics.

Comparing the MTS Helios and MTO Fusion Homoeopathics groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

t) MTO Izel and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-22: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Izel and MTS Homoeopathix.

Comparing the MTO Izel and MTS Homoeopathix groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

u) MTO Fusion Homoeopathics and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.063	.243	.155

Table 4-23: Mann-Whitney Test: Chemical Shift (δ) Values for MTO Fusion Homoeopathics and MTS Homoeopathix.

Comparing the MTO Fusion Homoeopathics and MTS Homoeopathix groups, the chemical shift values show a significant difference between all the peaks.

The hypotheses are thus accepted.

4.3 Relative Integration Values

4.3.1 Descriptives of relative integration values

STATISTICS=DEFAULT RANGE VARIANCE KURTOSIS SKEWNESS.

Valid cases = 36; cases with missing value(s) = 2.

Variable	*N*	*Mean*	*Std. Dev*	*Variance*	*Kurtosis*	*S.E. Kurt.*	*Skewness*	*S.E. Skew*	*Range*	*Min.*	*Max.*
H_2O	34	.92	.00	.00	-.39	.79	-.47	.40	.02	.91	.93
CH_2	34	.03	.00	.00	-.45	.79	.51	.40	.01	.03	.04
CH_3	34	.05	.00	.00	-.35	.79	.42	.40	.01	.04	.05

4.3.2 Kruskal-Wallis Test: Comparison of relative integration values

	H_2O	CH_2	CH_3
Significance (α)	0.000	0.000	0.000

Table 4-24: Kruskal-Wallis Test: Comparison of Relative Integration Values

The comparison of the relative integration values show a significant difference between the H_2O and CH_2 peaks for all the samples.

The hypotheses are thus accepted.

4.3.3 Mann-Whitney Test: Comparison of relative integration values

a) Control Lactose LM1 and MTO Garth

	H_2O	CH_2	CH_3
Significance (α)	.043	.024	.063

Table 4-25: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTO Garth

The comparison of the relative integration values of the Control Lactose LM1 and MTO Garth groups show a significant difference for the H_2O and CH_2 peaks.

The hypotheses are thus accepted.

The CH_3 peaks are not significantly different; hence the hypothesis for this value is thus rejected

b) Control Lactose LM1 and MTS Roy & Co.

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-26: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTS Roy & Co.

Comparing the Control Lactose LM1 and MTS Roy & Co. groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

c) Control Lactose LM1 and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.002	.000	.000

Table 4-27: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTS Helios.

Comparing the Control Lactose LM1 and MTS Helios groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

d) Control Lactose LM1 and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-28: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTO Izel.

Comparing the Control Lactose LM1 and MTO Izel groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

e) Control Lactose LM1 and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-29: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTO Fusion Homoeopathics

Comparing the Control Lactose LM1 and MTO Fusion Homoeopathics groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

f) Control Lactose LM1 and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-30: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTS Homoeopathix.

The comparison of the relative integration values of the Control Lactose LM1 and MTS Homoeopathix groups, show a significant difference between all the peaks.

The hypotheses are thus accepted.

g) MTO Garth and MTS Roy & Co.

	H_2O	CH_2	CH_3
Significance (α)	.018	.010	.026

Table 4-31: Mann-Whitney Test: Relative Integration Values for Control Lactose LM1 and MTS Roy & Co.

The comparison of the relative integration values of the MTO Garth and MTS Roy & Co. groups, show a significant difference between all peaks.

The hypotheses are thus accepted.

h) MTO Garth and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.001	.001	.002

Table 4-32: Mann-Whitney Test: Relative Integration Values for MTO Garth and MTS Helios.

Comparing the MTO Garth and MTS Helios groups, the relative integration values show a significant difference between all the peaks. The hypotheses are thus accepted.

i) MTO Garth and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.023	.013	.034

Table 4-33: Mann-Whitney Test: Relative Integration Values for MTO Garth and MTO Izel.

Comparing the MTO Garth and MTO Izel groups, the relative integration values show a significant difference between all the peaks. The hypotheses are therefore accepted.

j) MTO Garth and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.613	.713	.560

Table 4-34: Mann-Whitney Test: Relative Integration Values for MTO Garth and MTO Fusion Homoeopathics.

Comparing the MTO Garth and MTO Fusion Homoeopathics groups, the relative integration values do not show a significant difference on any of the peaks values.

The hypotheses are thus rejected.

k) MTO Garth and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.918	.792	.995

Table 4-35: Mann-Whitney Test: Relative Integration Values for MTO Garth and MTS Homoeopathix.

Comparing the MTO Garth and MTS Homoeopathix groups, the relative integration values do not show a significant difference on any of the peaks values.

The hypotheses are thus rejected.

l) MTS Roy & Co. and MTS Helios

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-36: Mann-Whitney Test: Relative Integration Values for MTS Roy & Co. and MTS Helios.

Comparing the MTS Roy & Co. and MTS Helios groups, the relative integration values show a significant difference between all peaks.

The hypotheses are thus accepted.

m) MTS Roy & Co. and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.022	.056	.012

Table 4-37: Mann-Whitney Test: Relative Integration Values for MTS Roy & Co. and MTO Izel.

Comparing the MTS Roy & Co. and MTO Izel groups, the relative integration values show a significant difference between H_2O and CH_3 peaks.

The hypotheses are thus accepted.

The CH_2 peak is not significantly different; hence the hypothesis for the value of this peak is rejected.

n) MTS Roy & Co. and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-38: Mann-Whitney Test: Relative Integration Values for MTS Roy & Co. and MTO Fusion Homoeopathics.

Comparing the MTS Roy & Co. and MTO Fusion Homoeopathics groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

o) MTS Roy & Co. and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-39: Mann-Whitney Test: Relative Integration Values for MTS Roy & Co. and MTS Homoeopathix.

Comparing the MTS Roy & Co. and MTS Homoeopathix groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

p) MTS Helios and MTO Izel

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-40: Mann-Whitney Test: Relative Integration Values for MTS Helios and MTO Izel.

Comparing the MTS Helios and MTO Izel groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

q) MTS Helios and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-41: Mann-Whitney Test: Relative Integration Values for MTS Helios and MTO Fusion Homoeopathics.

Comparing the MTS Helios and MTO Fusion Homoeopathics groups, the relative integration values show a significant difference between all the peak values.

The hypotheses are thus accepted.

r) MTS Helios and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-42: Mann-Whitney Test: Relative Integration Values for MTS Helios and MTS Homoeopathix.

Comparing the MTS Helios and MTS Homoeopathix groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

s) MTO Izel and MTO Fusion Homoeopathics

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-43: Mann-Whitney Test: Relative Integration Values for MTO Izel and MTO Fusion Homoeopathics.

Comparing the MTS Helios and MTO Fusion Homoeopathics groups, the relative integration values show a significant difference between all the peaks.

The hypotheses are thus accepted.

t) MTO Izel and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.000	.000	.000

Table 4-44: Mann-Whitney Test: Relative Integration Values for MTO Izel and MTS Homoeopathix.

Comparing the MTO Izel and MTS Homoeopathix groups, the relative integration values show a significant difference between all the peaks. The hypotheses are thus accepted.

u) MTO Fusion Homoeopathics and MTS Homoeopathix

	H_2O	CH_2	CH_3
Significance (α)	.025	.574	.076

Table 4-45: Mann-Whitney Test: Relative Integration Values for MTO Fusion Homoeopathics and MTS Homoeopathix.

Comparing the MTO Fusion Homoeopathics and MTS Homoeopathix groups, the relative integration values show a significant difference in the H_20 peak values.
The hypothesis is thus accepted.

The CH_2 and CH_3 peaks are not significantly different; hence the hypotheses for these peak values are rejected. The hypotheses are thus rejected.

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CHAPTER 5 - DISCUSSION

The objectives for this study, introduced in Chapter 1, were to:

- Compare and evaluate the nuclear magnetic resonance spectra of six samples of the homoeopathic remedy *Natrum muriaticum* LM1, representative of options available to homoeopathic practitioners in a medicinal potency (20% ethanol), to a Lactose LM1 control sample.
- Compare and evaluate the nuclear magnetic resonance spectra of these six known sources of the homoeopathic remedy *Natrum muriaticum* LM1 in medicinal potency (20% ethanol), to each other.
- Ascertain whether two samples produced with the same strict adherence to Hahnemann's' methodology, from same source materials and environmental conditions, would yield similar NMR Spectra, but distinct from a Lactose LM1 control sample.

5.1 First Objective

A summary of the results of Mann-Whitney statistical analysis in relation to chemical shifts and relative integration values (compared in terms of the CH_2, CH_3, and H_2O signals) which were detailed in Chapter 4, are presented below to facilitate further discussion.

5.1.1 Mann-Whitney Test: Chemical shift values for controls

Significance (α)	H_2O	CH_2	CH_3
Control Lactose LM1 - MTO Garth	.002	.072	.004
Control Lactose LM1 - MTS Roy & Co.	.000	.007	.004
Control Lactose LM1 - MTS Helios	.002	.000	.000
Control Lactose LM1 - MTO Izel	.000	.016	.000
Control Lactose LM1 - MTO Fusion Homoeopathics	.012	.002	.000
Control Lactose LM1 - MTS Homoeopathix	.188	.003	.000

Table 5-1: Mann-Whitney Test: Chemical Shift (δ) - Control

The purpose for the inclusion of the Lactose LM1 Control was to determine whether homoeopathic remedies would generate NMR spectra distinct from a similarly processed sample, where the crude substance was absent. Lactose is considered an inert substance in homoeopathy (Owen 2007: 265) and is a component within all the remedy samples as both solvent and substrate in preparation of LM potencies.

Any distinction in NMR spectra could be attributed to the presence of sodium chloride crude substance, since this is the key variable that differs between the control and the remedy samples. Both chemical shift and relative integration values of the CH_2, CH_3 and H_2O signals confirm that all homoeopathic *Natrum muriaticum* LM1 remedies were statistically unique in comparison to the LM1 Lactose control.

This is a desirable result, congruent with the source substance sodium chloride (NaCl) being responsible for the statistically significant differences

observed. Considering that dilution and form were identical among samples and the Control Lactose LM1 sample, representation of sodium chloride as a homoeopathic potency of *Natrum muriaticum* LM1 would be the singular distinction.

The lactose monohydrate sample used to manufacture the Lactose LM1 Control was the same batch utilised during trituration in both of the make-to-order (MTO) DUT samples (*Control Lactose LM1, MTO Garth & MTO Izel*). Lactose monohydrate is however a variable input during the manufacturing of the other remedy samples, which represents conditions prevailing within the marketplace.

The CH_2 peak in the '*Control Lactose LM1 - MTO Garth*' comparison was the only result not found to be statistically distinct from CH_2 peak in the Lactose control. For these samples only one batch of lactose monohydrate was used throughout both sample and control preparation, suggesting this could have been anticipated. However, the '*Control Lactose LM1 - MTO Izel'* samples shared this characteristic but were statistically distinctive. As both samples were also produced using same raw materials, crude substance, within the same laboratory environment and at the same time, technique emerges as a likely variable which may account for this result.

In addition, both remedy and control samples in this comparison (*Control Lactose LM1 - MTO Garth*) were produced by the researcher, so technique

and the action on the crude substance is a probable explanation for this distinction in the CH_2 peak.

Similarly, the H_2O peak in the '*Control Lactose LM1 - MTS Homoeopathix*' sample did not yield a statistically significant value.

While no conclusion can be drawn from these specific similarities, when NMR Spectra results are evaluated collectively, a fairly conclusive disparity between the samples and the Lactose LM1 control is clearly apparent. The statistically significant differences in the chemical shift values of the CH_2, CH_3 and H_2O signals collectively in these samples, implies that manufacturing practices do indeed play a role in the way physical properties inherent to the crude substance, *Natrum muriaticum* are developed, as was hypothesised.

5.1.2 Mann-Whitney Test: Relative integration values for controls

Significance (α)	**H_2O**	**CH_2**	**CH_3**
Control Lactose LM1 - MTO Garth	.043	.024	.063
Control Lactose LM1 - MTS Roy & Co.	.000	.000	.000
Control Lactose LM1 - MTS Helios	.002	.000	.000
Control Lactose LM1 - MTO Izel	.000	.000	.000
Control Lactose LM1 - MTO Fusion Homoeopathics	.000	.000	.000
Control Lactose LM1 - MTS Homoeopathix	.000	.000	.000

Table 5-2: Mann-Whitney Test: Relative Integration - Control

Relative Integration values of the CH_2, CH_3 and H_2O signals show that homoeopathic *Natrum muriaticum* LM1 remedies were unique and distinct

from the LM1 Lactose control. This is congruent with source substance sodium chloride (NaCl) being responsible for this statistically significant difference in all these samples. The relative areas below the each peak is created by the proportional signal strength detected, which is directly related to the number of nuclei and their relative arrangement at that that particular chemical shift value (Richards and Hollerton 2010).

The single CH_3 value in the "*Control Lactose LM1 - MTO Garth*" is intriguing as it may indicate that inexperience and technique somehow failed to develop this property fully. Again, since the researcher produced both the lactose control sample and the "*Control Lactose LM1 - MTO Garth*", perhaps some similarity should be expected in the resulting products, since this technique would be inherent to both sample and the LM1 Lactose control. When relative integration for this sample is analysed, the CH_3 value failed to reflect a statistically relevant value between these samples.

The "*Control Lactose LM1 - MTO Garth*" and "*Control Lactose LM1 - MTO Izel*" reduce the variables most completely, to just technique and the presence or absence of the sodium chloride crude substance. It may be that relative integration of CH_3 in particular reflects an element of technique, as all other samples are universally distinct. Unfortunately this remains conjecture, since so little is known about the nature of homoeopathic remedies or the significance of NMR spectra obtained in the context of ultra-diluted solutions. This is a primary constraint when attempting to interpret or extract any conclusion from results obtained.

The virtually universal statistically significant differences between relative integration values of the CH_2, CH_3 and H_2O signals collectively in these samples, suggests that manufacturing practices play a role in the physical properties measured, as hypothesised.

Certainly, when the chemical shift and relative integration results are considered globally, they suggest the existence of differences in physical structure, supporting the first hypothesis, which proposed that the *Natrum muriaticum* LM1 remedy samples will be distinctive in comparison to samples of an LM1 lactose control.

5.2 Second Objective

The Second Objective was to compare and evaluate the nuclear magnetic resonance (NMR) spectra between six sources of LM1 homoeopathic potency samples of *Natrum muriaticum* with one another, in terms of chemical shifts and relative integration values of the CH_2, CH_3 ,OH and H_2O signals, in order to determine if statistical differences existed, suggesting differences between their respective physical structures.

A summary of the results of Mann-Whitney statistical analysis in relation to chemical shifts and relative integration values (compared in terms of the CH_2, CH_3, and H_2O signals) which were detailed in Chapter 4, are presented below to facilitate further discussion.

5.2.1 Mann-Whitney Test: Chemical shift values for samples

Significance (α)	H_2O	CH_2	CH_3
MTO Garth - MTS Roy & Co.	.064	.031	.421
MTO Garth - MTS Helios	.002	.000	.000
MTO Garth - MTO Izel	.053	.374	.456
MTO Garth - MTO Fusion Homoeopathics	.000	.000	.000
MTO Garth - MTS Homoeopathix	.001	.000	.000
MTS Roy & Co. - MTS Helios	.088	.000	.000
MTS Roy & Co. - MTO Izel	.787	.062	.822
MTS Roy & Co. - MTO Fusion Homoeopathics	.000	.000	.000
MTS Roy & Co. - MTS Homoeopathix	.001	.000	.000
MTS Helios - MTO Izel	.074	.000	.000
MTS Helios - MTO Fusion Homoeopathics	.000	.000	.000
MTS Helios - MTS Homoeopathix	.000	.000	.000
MTO Izel - MTO Fusion Homoeopathics	.000	.000	.000
MTO Izel - MTS Homoeopathix	.000	.000	.000
MTO Fusion Homoeopathics - MTS Homoeopathix	.063	.243	.155

Table 5-3: Mann-Whitney Test: Chemical Shift (δ) – Nat mur LM1

In terms of chemical shift, the majority of samples revealed a statistically significant difference when compared to each other. This supports the second hypothesis that when variation in manufacturing a given homoeopathic remedy exists, evidence of differences will be reflected in results obtained from NMR investigation.

Even though the "*MTO Garth - MTO Izel*" samples showed no statistically significant difference in chemical shift after Mann-Whitney analysis, this degree of close similarity with respect to Chemical shift was anticipated and

outlined in the third hypothesis. Both were produced simultaneously, within the same environment and using the same raw materials (substances) and identical methodology. The purpose of producing these remedies to strict methodology was to reduce the number of variables to approximate any potential impact of technical experience and technique.

Confirmation that a specified approach using the same inputs and environment yielded the same chemical shift profile after NMR analysis is a fundamental requirement in order to test the second hypothesis and support the third hypothesis. An assertion that varied methodology would likely result in distinctive physical properties, discernible through NMR analysis has an implicit requirement that if this impact was negated, a similar NMR profile could be expected.

Consequently the lack of a statistically significant difference between these two samples supports the second hypothesis. It also raises questions related to the nature of the remedies themselves. Produced to conform more closely to Hahnemann's prescribed methodology, are they more representative therapeutically to the original provings? This falls out of this research scope of this investigation but remains an intriguing possibility which deserves further inquiry (Barthel 1993).

The close alignment between the *MTO - Garth* and *MTO – Izel* samples with the remedy sourced from Roy and Co. (India) in terms of chemical shift, would suggest a similar manufacturing methodology was used. Unfortunately this

could not be verified, as Roy & Co. could not conclusively trace the batch number of this sample back to their original supplier. This highlights another potential challenge practitioner's face in practice; where sourcing a remedy which proves really useful within their practice may not always be possible and could potentially impact the quality of their inventory and consistency of their clinical results.

It is acknowledged that it is highly probable that this *MTS Roy & Company* sample was manufactured using distinctly different methodology. If this indeed was to be the case, varied methodology would have been shown to yield statistically identical NMR spectra in terms of the CH_2, CH_3 and H_2O signals. The second hypothesis of this study would not have been met in this case.

Another anomaly is presented when comparing the chemical shift Mann-Whitney results for the *MTO - Fusion Homoeopathics* and *MTS – Homoeopathix* samples. The *MTO - Fusion Homoeopathics* remedy was manufactured specifically for this study using exactly the same source substance used in the *MTO - Garth* and *MTO – Izel* samples. Both companies share a similar geography and climate, but their manufacturing principles differ considerably, as outlined in their manufacturing documents. (See Appendix B)

In addition, the crude substance source is not identical and while *MTO - Fusion Homoeopathics* remedy is produced using ozonated water, the *MTS - Homoeopathix* sample is produced with distilled water. It was expected that

the distinction between the methodologies and manufacturing philosophy for these remedies would be reflected on their respective NMR spectra. This however, was not the case. The comparison between these samples does not support the second hypothesis.

The *MTO - Fusion Homoeopathics* remedy was produced by a highly experienced specialist practitioner, who produced the remedy within an environment designed to conform to good manufacturing practice (GMP) guidelines, according to 'HAB7' procedures (see Appendix B), so a more analogous result with the remedies produced at DUT laboratory was expected. Again, this was not the case and this sample was distinctive in all measured peaks to the *MTO - Garth* and *MTO – Izel* samples. Again this is inconsistent with the second hypothesis.

5.3 Third Objective

Significance (α)	H_2O	CH_2	CH_3
MTO Garth - MTO Izel	.053	.374	.456
MTS Roy & Co. - MTO Izel	.787	.062	.822

Table 5-4: Mann-Whitney Test: Chemical Shift Table Extract - Nat mur LM1

Significance (α)	H_2O	CH_2	CH_3
MTO Garth - MTO Izel	.023	.013	.034

Table 5-5: Mann-Whitney Test: Relative Integration Table Extract - Nat mur LM1

The "*MTO Garth - MTO Izel*" samples were produced simultaneously, within the same environment and using the same raw materials (substances) and identical methodology. The purpose of producing these remedies to strict methodology was to reduce the number of variables to approximate any potential impact of technical experience and technique. The absence of a statistically significant difference in chemical shift after Mann-Whitney analysis supports the third hypothesis.

The relative areas below the each peak is created by the proportional signal strength detected, which is directly related to the number of nuclei and their relative arrangement at that that particular chemical shift value (Richards and Hollerton 2010). Consequently the statistically significant differences in all the peaks analysed between the "*MTO Garth - MTO Izel*" samples may be indicative of the degree to which technique elicits molecular orientation at those points measured. Certainly, at face value these results do not support the third hypothesis.

An alternative interpretation is that even when the variables related to remedy production on the LM scale are minimised, this does not necessarily translate to predictable NMR spectra profiles. Clinical success with remedies from multiple sources suggests that the molecular dynamics under scrutiny during NMR analysis do not correlate with the therapeutic nature of importance clinically.

5.4 Practical Considerations

It would appear that NMR spectra may not be an appropriate technology for application in quality monitoring or quality control of homoeopathic remedies. This may be a restriction limited to low alcohol concentrations, so may yield more encouraging results when analysis of this nature is performed on medicinal potency alcohol concentrations. Fundamentally though, until there is a comprehensive understanding of the significance and role of the chemical shift and relative integration of the specific peaks, we have no way of ensuring quality control of any practical value clinically, with this technology.

It may be that Relative Integration in LM remedy production may be influenced during the trituration or succussion stages. During trituration as the lactose and crude substance is ground between mortar and pestle, substantial energy is likely transferred to develop the remedy in some way. Perhaps experience (technique) then is a core variable, which would validate Hahnemann's meticulous approach to technique and method. Technique is most apparent during trituration, where consistency and nature of energy transfer are likely to play an integral role in developing the C3 Triturate used to produce the LM Mother Tincture (LM0) as detailed in Appendix A.

Succussion is another variable where the force distribution could be influenced by technique, though one would expect this to play a larger role in remedies where succussion represents a more profound activity in

manufacture of the remedy. In the footnote to aphorism 270, Hahnemann warns that in other potencies which are highly succussed, dangerous and undesirable changes can result in remedies (Hahnemann and O'Reilly 2001). Hahnemann believed even a small number of succussions of the LM patient prescription would alter and develop and intensify the potency, as he explains in aphorism 248 of his 6th edition *Organon* (Hahnemann and O'Reilly 2001).

The distinctive action of LM potencies is believed to be due to the relatively low degree of succussion. The Helios manufacturing document refers to a drop of 3C, implying serial dilution, which may explain this sample's largely distinct nature in relation to other remedies, except some similarity of H_2O chemical shift to *"MTS - Roy & Company"* and *"MTO – DUT – Izel samples".*

Each LM potency increase represents 1:50,000, approximately 2.5 on the centesimal scale (CH) (Morgan and Helios Pharmacy 2014). As the LM mother tincture is produced at 3C, LM 1 then would roughly approximate to 5.5CH, marginally below the molar threshold limitation of Avogadro's number (6CH). What becomes evident is how easily this value is affected by even marginal deviation from perfect methodology.

Perhaps this impact could be considered as fairly inconsequential, especially given the subsequent process of patient succussion, which would change the remedy anyway. On the other hand, a lack of professionalism in manufacturing practices could have unanticipated effects therapeutically, especially on sensitive individuals.

The sample produced by the more experienced Homoeopharmaceutics Senior Lecturer, Dr Izel Botha ("*MTS Helios - MTO Izel"*), revealed no statistical difference, while "*MTO Garth - MTS Helios"* analysis was statistically distinct for all measured points. If relative integration is indeed a reflection of the relative symmetry within the molecular lattice (Richards and Hollerton 2010), influencing the intensity of the NMR signal detected between samples analysed, then technique again emerges as the core variable. It is likely that improved technique of the more experienced Homoeopharmaceutics senior lecturer resulted in distinctive relative integration values in her samples, which may be due to an improved ability to induce more consistent energy transfer during trituration and or succession steps, since dilution is identical.

NMR spectra peaks were at the same position for the "*MTO Garth - MTO Izel*" samples, but had different areas below their curves. This implies that when variables are reduced to human technique and the "fingerprint" (Blümich 2005) of chemical shift is attained, a human element still plays a role in the physical properties attained reflected in properties inherent to relative integration.

5.4.1 Mann-Whitney Test: Comparison of relative integration values

Significance (α)	H_2O	CH_2	CH_3
MTO Garth - MTS Roy & Co.	.018	.010	.026
MTO Garth - MTS Helios	.001	.001	.002
MTO Garth - MTO Izel	.023	.013	.034
MTO Garth - MTO Fusion Homoeopathics	.613	.713	.560
MTO Garth - MTS Homoeopathix	.918	.792	.995
MTS Roy & Co. - MTS Helios	.000	.000	.000
MTS Roy & Co. - MTO Izel	.022	.056	.012
MTS Roy & Co. - MTO Fusion Homoeopathics	.000	.000	.000
MTS Roy & Co. - MTS Homoeopathix	.000	.000	.000
MTS Helios - MTO Izel	.000	.000	.000
MTS Helios - MTO Fusion Homoeopathics	.000	.000	.000
MTS Helios - MTS Homoeopathix	.000	.000	.000
MTO Izel - MTO Fusion Homoeopathics	.000	.000	.000
MTO Izel - MTS Homoeopathix	.000	.000	.000
MTO Fusion Homoeopathics - MTS Homoeopathix	.025	.574	.076

Table 5-6: Mann-Whitney Test: Relative Integration – Nat mur LM1

This of course predisposes an assumption that the physical properties apparent during NMR analysis are in fact relevant to the nature and effect of a homoeopathic remedies action demonstrated in clinical practice. Certainly this is plausible, as the physical properties of ultra-diluted solutions have repeatedly been shown to exhibit distinct and unexpected features during laboratory investigations, using many laboratory and technological approaches, some of which were mentioned in Chapter 2 - *Other Potential Technologies*.

A medicinal potency (alcohol content of 20%) was chosen to simulate “real world” application as prescription to patients, based on a previous study where a potency of 15% produced promising (Erasmus 2004). It would appear that the H_2O peak may have been misidentified as an OH peak in this study. The absence of a distinctive OH peak on NMR spectra in this study is attributed to the low alcohol percentage used, since studies undertaken at medicating potency alcohol concentrations (87% and 95% in previous DUT NMR studies) all feature prominent OH peaks on NMR spectra.

Consequently, only the H_2O, CH_2 and CH_3 produced peaks which facilitated processing and statistical analysis in this study. Dr J Brand ascribed the absence of an OH peak to the alcohol concentration being too low in this study as the miscibility of alcohol and technical sensitivity of NMR Spectrometer require a far higher alcohol concentration for a significant OH peak to materialise on NMR Spectra. This represented an error in the study design philosophy, since a stated intention was to build on the existing body of work undertaken using NMR technology, while this anomaly and reliance on CH_2, CH_3 and H_2O spectra peaks does not conform to that objective.

Further, despite randomising the samples for analysis, the lactose and alcohol controls were easily identified during NMR analysis. The appearance and odour characteristics for these samples set them apart from the other samples for analysis. As these controls were not potentised and served primarily as reference substances and quality control, they were not required to test any of the hypotheses and so did not impact or bias the results.

Reusing DSMO capillary tube is standard practice during NMR analysis, due to the expense of this reagent. However, handling and environmental exposure during transfer across the 50 samples analysed, logically introduces a significant risk of contamination in the context of high dilution analysis. This is an aspect of methodology which was not anticipated ahead of analysis and an aspect which the researcher would approach attempt to better mitigate during analysis.

Despite the vast range of physical investigations that have been directed towards investigating homoeopathic preparations (some of which were highlight in chapter 2), consensus regarding the nature and significance of this type of research, has not materialised (Cartwright 2015).

Hahnemann's meticulous attention to detail, lengthy evolution in remedy production and his aversion to even the slightest deviation to remedy manufacturing from his proven method producing LM potency, discussed in Chapter 2, may have been well-founded. Actual manufacturing practises which claim to follow the 6th edition *Organon*, fail to do so exactly and agreed standards outlined in the various Pharmacopeia entrench this deviation consistently. Deviation from that method towards a consensus approach to manufacturing could have unintended consequences.

It seems plausible that the pursuit of very specific outcomes in clinical practise (Similimum) and in research (repeatability, validity) could be undermined by vague and inconsistent manufacturing processes. A key objective motivating this research was the potential practical implication for practitioners in clinical practise, who interface with patients. Ultimately research should align to patient interests, since this NMR study reflects a significant variation in the physical properties between potential sources of supply; this would likely be of concern to Hahnemann.

Until an agreed technology is able to reliably represent identified therapeutic properties inherent in homoeopathic remedies, reliability, consistency and caution are likely to be prudent principles in practice. In reality, manufacturing processes do not necessarily align with practices used for the remedies that were taken during provings or used in the clinical cases which are included in references like the *Materia Medica* and *Repertories*.

If we assume remedy manufacturing to play a role in the nature of the remedy expression, surely it is likely that inconsistency is ill-advised as the results of this NMR study would suggest. This study however does not presume that NMR properties reflected on NRM spectra are explicitly relevant to the therapeutic properties of significance. On the contrary, despite manufacturing deviation from the prescribed methodology, practitioners and patients routinely witness and experience therapeutic benefits. This raises concern regarding the relevance of the dimensions scrutinised during NMR analysis. Especially considering the environment remedies are subjected to during analysis.

There is no intention, explicit or implied to criticize or compare suppliers with an assertion that a conclusion can be derived related to suitability, quality or therapeutic value from the NMR analysis data derived. This investigation is limited to merely processing remedies through NMR to discern spectra differences and highlight material and methodology as potential variables for the results attained.

Certainly, other variables could account for these results, though the spectra obtained during this study, together with the body of work preceding and lying ahead is important and prerequisite to better understanding the nature of ultra-diluted solutions and homoeopathic remedies in particular, there is currently no way to correlate the NMR spectra from analysis to the therapeutic implications for a given remedy.

CHAPTER 6 - CONCLUSION AND RECOMMENDATIONS

6.1 Conclusions

Natrum muriaticum LM1 samples sourced from a variety of make-to-order (MTO) and make-to-stock manufacturing environments produced almost universally distinct NMR spectra profiles in terms of chemical shifts and relative integration values of the CH_2, CH_3 and H_2O signals, at an alcohol concentration of 20%, when compared to a Lactose LM1 control sample, directly supporting the first hypothesis.

When the NMR spectra of make-to-order (MTO) and make-to-stock (MTS) remedies were analysed and compared to each other in terms of chemical shifts and relative integration values of the CH_2, CH_3 and H_2O signals, they were for the most part statistically distinct, but some anomalous results emerged. No feature inherent to the methodology that would account for these similarities could be identified; indeed by all accounts there methodology was dissimilar. This raises concern related to the nature and importance of the molecular changes being scrutinised using NMR to the remedy development.

There was no statically significant difference in NMR spectra in terms of chemical shift, between the two samples produced in the same environment using the methodology to closely aligned to the *Organon* (Hahnemann and

O'Reilly 2001), yet in terms of relative integration values, they were statistically distinct. This may be the impact of technique, where method and material correlate with a remedy identity on NMR spectra, but remedy development is translated differently with regards to molecular orientation.

Homoeopathic research is frustratingly devoid of certainty or definitive conclusion, here it is not possible to identify the variables or account for the nature of NMR spectra that are produced after sample analysis. This study concludes that while NMR was able to discern physico-chemical distinction between LM1 potency *Natrum muriaticum* remedies to control conclusively, this does not translate to practical application of NMR for quality control or remedy comparison for LM potencies at alcohol content levels, associated with medicinal prescription.

Given that remedies produced to strict protocol resulted in statistically equivalent product is very encouraging, however distinct anomalies within the results suggest that this technology may not present a reliable platform for this investigation LM potencies as a patient prescription of 20% alcohol.

6.2 Recommendations

It is imperative though that these studies be repeated to investigate the reproducibility of the results. Some improvements for consideration in future NMR studies that resemble the objectives undertaken here:

- Fully blinded
    - The requirement introduced with this study to undertake the physical NMR analysis personally, while well-intentioned, introduces the risk of bias where controls are easily identifiable from ultra-diluted solutions.
- Smaller scope more runs
    - Study design may consider the analysis of fewer samples for analysis, which would allow larger sample sizes within the limited budget available.
    - Small sample size of only 5 readings per sample, was determined by financial constraints rather than an ideal sampling ratio for study design
- T1/T2 relaxation relaxation time studies are potentially more sensitive and better suited to NMR analysis of homoeopathic dilutions.
- Given the limitations which emerge translating NMR spectra into practical understanding of the nature of the variables underlying those results, perhaps investigation using some of the new alternative technologies may be considered.

- An investigation to determine the minimum alcohol percentage in homoeopathic dilutions that would allow NMR analysis to be comparable with the bulk of the NMR research already undertaken by DUT may be of value.
- It would appear that during homoeopathic research, methodology for remedy production should be standardised, or sourced from one reliable source where methodology is known.
    - This study would suggest that since remedies reveal subtle differences even when variables are reduced to the person making the remedy (as is the case with the MTO DUT remedies), research remedies should be sourced from a known quantity. Manufacturing documents and quantities used and methodologies are known.
- It may be useful to compare remedies produced mechanically versus manually for LM potency to investigate the impact of technique on NMR spectra in isolation.

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APPENDICES

Appendix A - Hahnemann and LM Methodology

The methodology meticulously detailed by Hahnemann in aphorism 270, of his 6th edition *Organon* (Barthel 1991; Hahnemann and O'Reilly 2001; Jütte 2007) is the realisation of his life's work, so with deep respect is used as a basis for the following outline of his method.

1CH Trituration

During LM preparation substances are triturated to 3CH, at which point even metals become soluble.

- One troy grain of powdered substance is added to mortar
- Trituration
    - Each trituration is divided into three stages, initiated by adding first third of 100 grains of lactose, mixed with contents of mortar
        - Trituration with pestle for 6-7 minutes
        - Mortar scraped for 3 – 4 minutes to form a homogeneous mixture
        - Trituration with pestle for 6-7 minutes
        - Mortar scraped for 3 – 4 minutes to form a homogeneous mixture
    - Next third of 100 grains added and process repeated
    - Last third of 100 grains added and process repeated

2CH Trituration

- one grain of the powdered 1CH Trituration is added to mortar
- Trituration as per 1CH methodology

3CH Trituration

- one grain of the powdered 2CH Trituration is added to mortar
- Trituration as per 1CH methodology

Note:

Each of the three stages takes 20 minutes to complete, so each stage of trituration lasts an hour, hence 3 hours to complete trituration.

LM Mother Tincture

- One grain of 3CH
- Then dissolved in 500 drops of mixture (1 part alcohol: 4 parts distilled water), by gentle rotation about the vials axis (lactose able to dissolve in this alcohol concentration)

LM1

- One drop is transferred to a vial and 100 drops of alcohol added, where resulting level in vial is 2/3 full
- 100 hard succussions against a hard, but elastic surface

0/1 Granules

- 500 Starch / sugar granules used of a size where approximately 100 granules weighed a grain.
- These where saturated using drops of the LM1, then quickly spread on blotting paper to dry

LM2

- One granule of 0/1 was allowed to dissolve in 1 drop of distilled water, then 100 drops added, where resulting level in vial is 2/3 full
- 100 hard succussions against a hard, but elastic surface

0/2 Granules

- 500 Granules used where approximately 100 granules weighed a grain.
- These where saturated using drops of the LM02, then quickly spread on blotting paper to dry

Further potencies

- Prepared similarly, using liquid from the preceding potency, to impregnate granules providing maximum surface area to volume (Owen 2007).

Appendix B - Preparation of Natrum muriaticum LM (0/1)

(Adapted From GHP, 1985 and Hahnemann, 1991)

(German Homoeopathic Pharmacopoeia 1985)

Apparatus:

Unglazed porcelain pestle and mortar

Steel spatula

Mass balance (calibrated & accurate)

Lighter

96% alcohol (svr)

Clean, empty vials

Filter paper

Labels

Lactose BP powder

Natrum muriaticum (Laboratory grade)

25ml Amber Glass Bottle (AGB)

Pasteur pipette - Borosilicate (7x)

Neoprene bulb

90% ethanol

Distilled water (Aq Dist)

5ml screw-top bottle

Labels

Note:

Centesimal (1: 100) ∴ Lactose: 3 x 3,3g = 9.9g / *Natrum muriaticum* 0.1g

Method Overview:

Environment, apparatus and utensils must be clean and odourless

Mortar, pestle and spatula:

- Clean with distilled water, and flame using 96% alcohol (and lighter).
- Allow to cool before use.

Tare a new piece of filter paper on mass balance.

Mass 0.1 g of *Natrum muriaticum* on filter paper.

Tare a new piece of filter paper on mass balance (repeat this step 3x)

Mass 3.3g of pure lactose powder onto filter paper.

1CH Nat-mur Triturate

Place 3.3g of lactose into mortar and triturate for a short period.

Add the 0.1g crude *Natrum muriaticum* to mortar.

Triturate for 6 minutes → scrape down for 4 minutes using spatula. (Repeat this step) ∴ total trituration time ≡ 2 x 10 minutes = 20 minutes

Add the second portion of 3.3g of lactose powder. (Repeat above step)

Add final 3.3g of lactose powder. (Repeat above step)

Total trituration time for 1CH ≡ 20 minutes x 3 = 1 hour

Decant 1CH triturate into a vial and label "*Natrum muriaticum* 1CH".

2CH Nat-mur Triturate

Repeat steps for 1CH, but substitute 0.1g *Natrum muriaticum* 1CH (for *Natrum muriaticum* crude substance).

Decant 2CH triturate into a vial and label "*Natrum muriaticum* 2CH".

3CH Nat-mur Triturate

Repeat steps for 2CH, but substitute 0.1g *Natrum muriaticum* 2CH (for Natrum 0.1g *Natrum muriaticum* 1CH).

Decant 3CH triturate into a vial and label "*Natrum muriaticum* 3CH".

Nat-mur LM Mother Tincture (LM0)

Select a 50ml Amber Glass Bottle (AGB)

Tare a new piece of filter paper on mass balance

Mass 0.065g (1 Troy grain) of *Natrum muriaticum* 3CH triturate on filter paper

As 500 Troy grains is equivalent to 32,426 ml (1 part ≡ 0.0648g), 4 parts AqDist = 25.941ml plus 1 part ethanol (90%) = 6.485ml.

Decant 1 Troy grain (0.065g) of 3CH triturate into AGB, add 25.941ml Aq Dist and swirl gently around axis of the bottle to dissolve completely (Do not succuss, swirl gently to mix).

Add 1 part ethanol (90%) = 6.485ml.

This would represent LM mother tincture (LM0)

Label sample appropriately as "LM0"

LM1 Medicating Potency

Into a 10ml Amber Glass Bottle (AGB)

- Add 1 Troy grain of LM0 mother tincture (0.065g)
- Add 6.485ml ml of 95% ethanol into 10ml Amber Glass Bottle (AGB)

Succuss 100x

Label sample appropriately as “LM1”

LM1 (0/1) Granules

Measure out 1000 granules ≡ 1.717g of granules into 50ml beaker

Add 0.13ml of LM1 medicating potency to saturate the granules

Swirl and agitate granules to mix and dry

Decant into clean 5ml screw-top bottle

Label sample appropriately as Label sample appropriately as 0/1

Note: This LM1 label will be substituted with a random reference number prior to NMR analysis to eliminate any bias.

B.1 MTO - Fusion Homoeopathics cc - Triturations

Fusion Homoeopathics CC

Unit B4, Micro Industrial Park, 17-19 Hammer Ave,
Strydom Park, 2169 Tel. 011 027 3665

MASTER MANUFACTURING DOCUMENT

Document version nr: 001 **Effective date: March 2012**

MANUFACTURING PROTOCOL FOR **TRITURATIONS**

PRODUCT NAME:	Natrium chloratum - Garth Miller		
Manufacturing method:	**HAB 6**	**BN Starting Material:**	not given
BATCH NO:	G13045	**EXPIRY DATE:**	
DATE COMMENCED:	16/07/2013	**DATE COMPLETED:**	23/07/2013

MANUFACTURING METHOD: Room temp. 14°C

Instructions	Checked by
1. Ensure working area is clean and correct.	
2. Collect the required number of containers and lids.	
3. Weigh off ~~0,1g~~ of the starting material. 0,3g	
4. Weigh off ~~3 x 3,3~~g of lactose. 3 x 9,9g	
5. Add the starting material and ~~9~~,9g of the lactose into the mortar.	
6. Triturate the combination for 6 minutes.	
7. Scrape the sides of the mortar for 4 minutes.	
8. Repeat steps 6 & 7.	
9. Add a further 9,9g of lactose and repeat steps 6 – 8.	
10. Add the last amount of 9,9g of lactose and repeat steps 6 – 8.	
11. Transfer the entire contents into an appropriate container and label as **1CH Trit.**	
12. Weigh off 0,3g of the **1CH Trit.**	
13. Weigh off 3 x 9,9g of lactose.	
14. Add the 1CH Trit and 9,9g of the lactose into the mortar.	
15. Triturate the combination for 6 minutes.	
16. Scrape the sides of the mortar for 4 minutes.	
17. Repeat steps 15 & 16.	
18. Add a further 9,9g of lactose and repeat steps 15 – 17.	
19. Add the last amount of 9,9g of lactose and repeat steps 15 – 17.	
20. Transfer the entire contents into an appropriate container and label as **2CH Trit.**	

Master Copy Certified Correct:

Document code:	MD-12C01-TRIT01	Revision no.:	1	Effective date:	26.03.2012
Supercedes no.:	N/A	Effective date:	N/A	Reason for update:	New document
Prepared by:	U. HOHL		Date:	26.03.2012	
Head Disp. Lab:			Date:	26.03.2012	

Instructions	Checked by
21. Weigh off 0,3g of the **2CH Trit.**	
22. Weigh off 3 x 9,9g of lactose.	
23. Add the 2CH Trit and 9,9g of the lactose into the mortar.	
24. Triturate the combination for 6 minutes.	
25. Scrape the sides of the mortar for 4 minutes.	
26. Repeat steps 24 & 25.	
27. Add a further 9,9g of lactose and repeat steps 24 – 26.	
28. Add the last amount of 9,9g of lactose and repeat steps 24 – 26.	
29. Transfer the entire contents into an appropriate container and label as **3CH Trit.**	
30. Store all other bottles below 25°C and out of direct sunlight.	

[redacted]

Master Copy Certified Correct:

Document code:	MD-12C01-TRIT01		Revision no.:	1	Effective date:	26.03.2012	
Supercedes no.:	N/A	Effective date:	N/A		Reason for update:	New document	
Prepared by:	U. HOHL			Date:	26.03.2012		
Head Disp. Lab:				Date:	26.03.2012		

B.2 MTO - Fusion Homoeopathics cc - LM Potencies

Fusion Homoeopathics CC Unit B4, Miero Industrial Park, 17-19 Hammer Ave, Strydom Park, 2169 Tel. 011 027 3665

MASTER MANUFACTURING DOCUMENT

Document version nr: 001 **Effective date: July 2013**

MANUFACTURING PROTOCOL FOR CONVERSION OF TRITURATIONS TO LM POTENCIES

PRODUCT NAME:	Natrum muriaticum LM 1 - G. Miller		
Manufacturing method:	**HAB 17**	**SN Starting Material:**	G13045
BATCH NO:	G13072	**EXPIRY DATE:**	07/2016
DATE COMMENCED:	25/07/2013	**DATE COMPLETED:**	26/07/2013

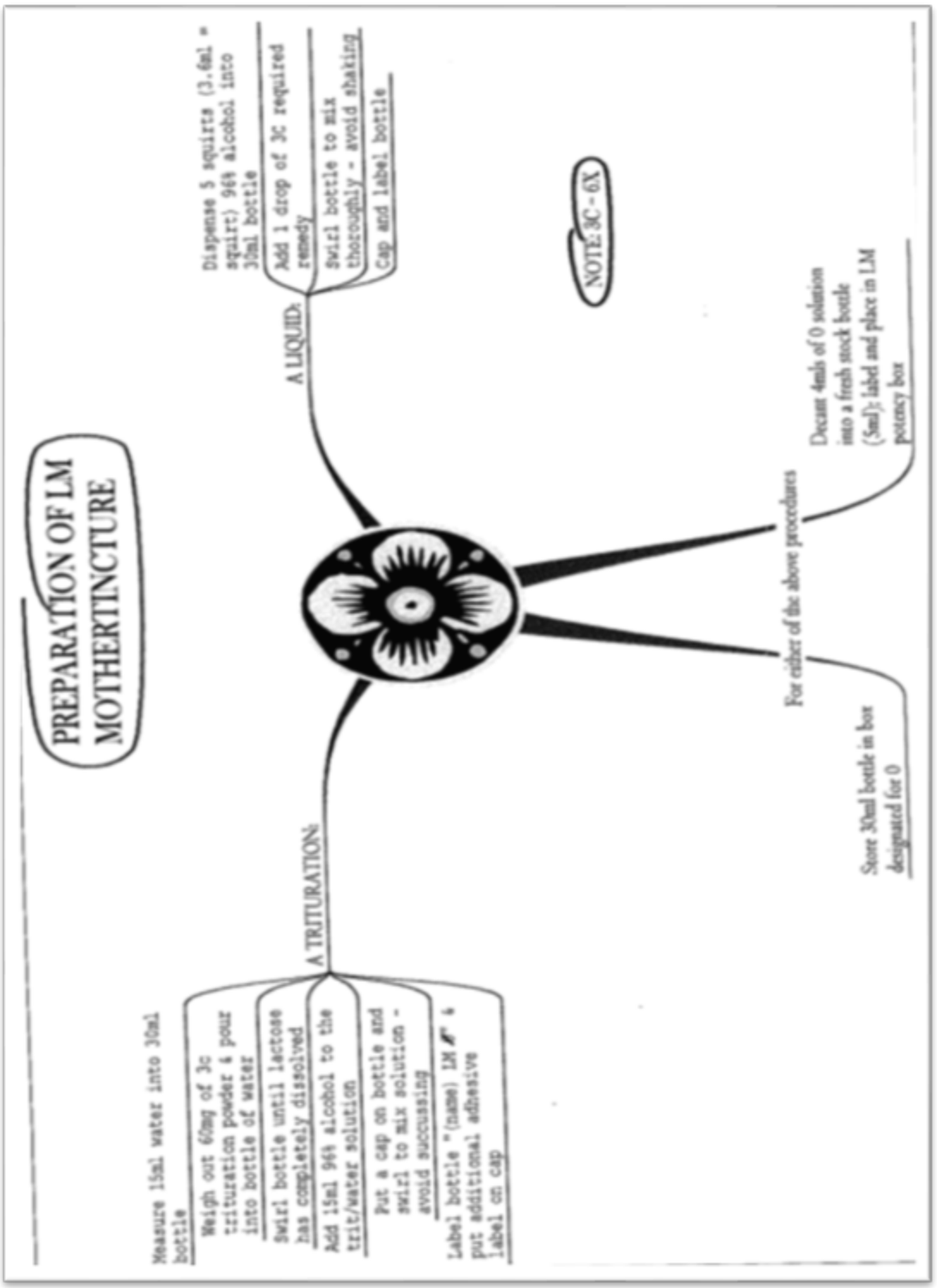
MANUFACTURING METHOD:

Instructions	Checked by
1. Ensure working area is clean and correct.	AM
2. Collect the required number of containers and lids.	AM
3. Weigh off 0,6g of the 3C Trit.	AM
4. Add the 0,6g Trit to 200mL of ethanol 15% (*m/m*).	AM
5. Ensure that the Trit dissolves completely.	AM
6. Add 1 drop of this solution to 2,5mL of ethanol 86% (*m/m*).	AM
7. Close the bottle tightly and succuss 100 times.	AM
8. Use this solution to consistently impregnate 100g of Size 10 globules.	AM
9. Allow these granules to air dry.	WA
10. Transfer the entire contents into an appropriate container and label as **LM 1.**	WA
11. Collect the required number of containers and lids.	
12. Dissolve 1 globule of the LM 1 in 1 drop of purified water.	
13. Add 2,5mL of ethanol 86% (*m/m*).	
14. Close the bottle tightly and succuss 100 times.	
15. Use this solution to consistently impregnate 100g of Size 10 globules.	
16. Allow these granules to air dry.	
17. Transfer the entire contents into an appropriate container and label as **LM 2.**	

Master Copy Certified Correct:

Document code:	MD-13G01-LM01		Revision no.:	1	Effective date:	23.07.2013
Supercedes no.:	N/A	Effective date:	N/A		Reason for update:	New document
Prepared by:	U. HOHL			Date:	23.07.2013	
Head Disp. Lab:				Date:	23.07.2013	

B.3 MTS – Homoeopathix Manufacturing Document



PREPARATION OF LM STOCK

PREPARATION OF LM 1 FROM LM 0

- Dispense 1 squirt (3.6ml) of 96% alcohol into amber stock bottle
- Add 1 drop of LM 0
- Cap bottle and succuss 100 times. Label "LM 1"
- Place bottle in the box next to the LM 0

Apart from the preparation of LM 1, all LMs are prepared by medicating a single size 0 sucrose granule with the preceding potency

PREPARATION OF HIGHER LMs FROM LM1 & ABOVE

- Lay a 5cm filter paper which has been creased, flat onto a clean paper towel
- Carefully dispense onto this a single "poppy seed" LM granule so that it rests in the crease
- Drop 1 drop of the preceding LM potency onto the granule, ie if you are making LM 3 drop 1 drop on LM 2 onto the granule
- Take a fresh glass vial and squirt a few drops of purified water into the bottom. Shake the water out of the vial so that only the merest drop remains in the bottom
- Carefully lift the filter paper with moistened granule & drop the granule into the vial. If the granule clings to the inside wall of the vial, tap the tube against the counter top to dislodge it so that it falls into the droplet of water at the bottom of the vial. Cap vial and label.
- leave the vial for about 5 minutes to allow the granule to dissolve in the water
- After a few minutes check to see if granule has dissolved. gently tapping or flicking the bottom of the vial with your finger can help to break up the granule.
- Once it is dissolved, dispense one squirt of 96% alcohol into a fresh vial. Pour the alcohol in this vial into the first vial with the dissolved granule.
- Replace the cap and succuss 100 times

NOTE: 3C - 6X

MAKING LMs FROM CENTESIMAL POTENCIES ABOVE 3C

- Where the lowest potency is a 4C or 5C:
  - Dilute 1 in 5 to get LM 0, ie into 1 squirt of 96% alcohol add 0.9ml of 4C. Mix but do not succuss
  - From 5C: Dilute 1 in 5 to get LM1, ie into 1 squirt of 96% alcohol, add 0.9ml of 5C & succuss 100 times. There is no LM 0 in this case
- From 6C: the lowest available is a LM 2
  - From 6C: Dilute 1 in 500 (ie 1 drop to 5 squirts of 96% alcohol) & mix but do not succuss. Then dilute 1 in 5: 0.9ml into 1 squirt of alcohol & succuss 100 times. This will give LM 2. There is no LM 0 or LM 1 in this case. The bottle of 1 in 500 should be labelled "LM 1 PLUS" & kept in a separate bank in order not to confuse these with our regular LM 0s.

TO PREPARE 2GRAM VIALS OF "POPPY SEED" GRANULES MEDICATED WITH LM POTENCIES OF MEDICATED ALCOHOL

1. Take a plastic tray & wipe clean with alcohol and place 2 squares of paper towel, folded in half & placed side by side in tray to line bottom

2. Take one 15ml filter paper & fold it in half twice & write the name & LM number near the curved edge of the folded paper

3. Open this into a cone & peg this by its edge on the ring on the ring stand

4. Pour one vial-full of 'poppy seed" granules into the cone

5. Drop 10 - 12 drops of LM potency onto the granules. Let this stand for a few minutes to allow the alcohol to flow through the granules. Make sure the alcohol on the granules does not soak upward on the filter paper to the point where it contacts either the clothes peg or metal ring, otherwise there is a danger of contaminating subsequent remedies

6. Remove the cone from ring, collapse the cone (fold flat), fold the edge down & then shake the granules around lightly to ensure that all granules are thoroughly medicated

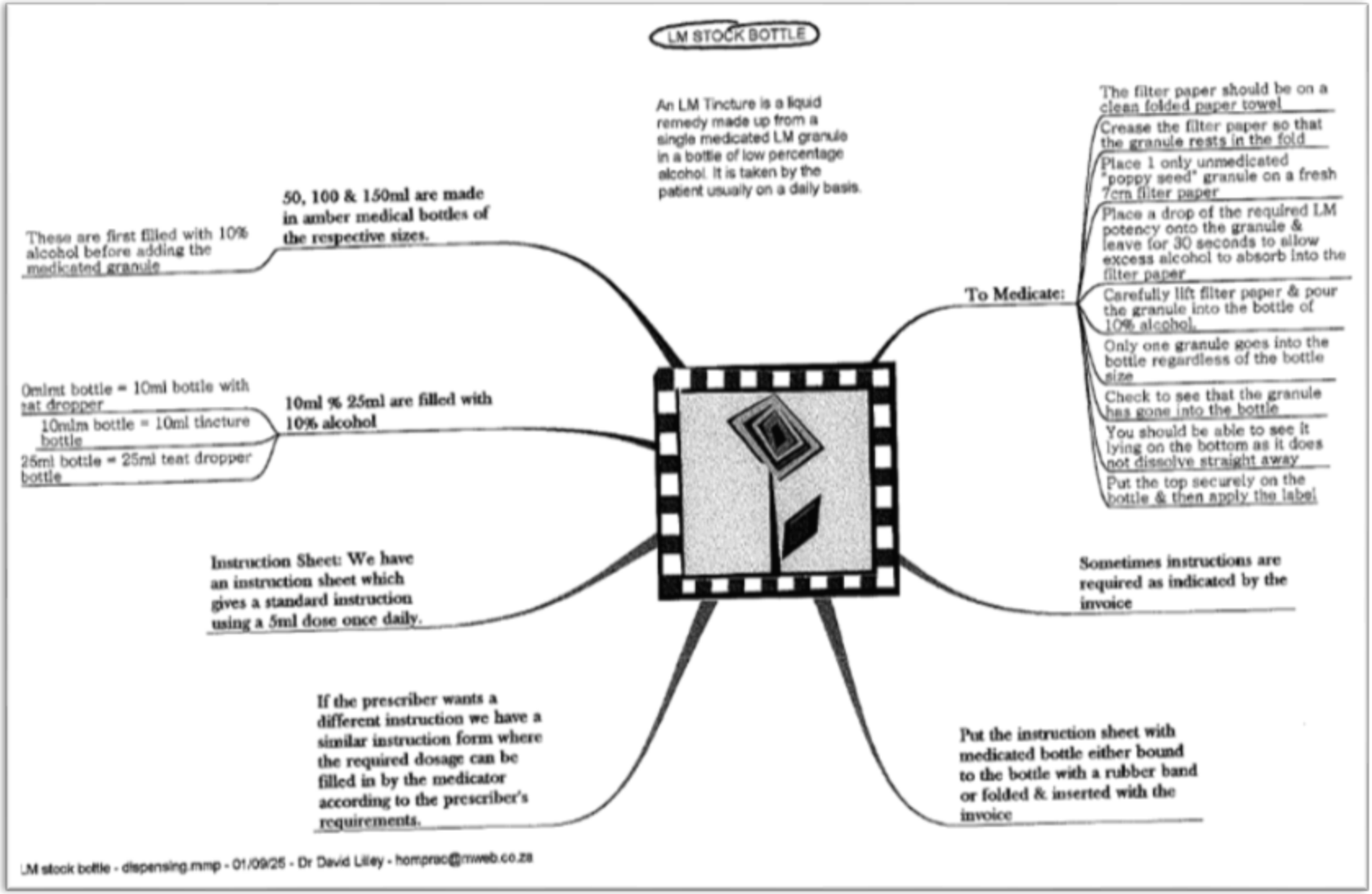
7. Place the folded cone flat onto the paper towel in tray with the remedy name at the edge of the paper exposed to view

8. Continue preparing LMs in this fashion, placing them in the tray ensuring that they are well away from each other to prevent cross-contamination. The maximum number of LMs to a tray is 4

9. Allow these to dry thoroughly. Approx. 1/2 hour in summer

10. Once LMs are dry they are individually lifted from the trays, & the filter paper partially unfolded & carefully pressed by hand to break up the granules which have now clumped during the drying process. The granules are then poured into 2 gram vials, the plastic tops are put on & they are each labelled.

The granules are used by practitioners to make up LM tinctures for their patients


LM STOCK BOTTLE
An LM Tincture is a liquid remedy made up from a single medicated LM granule in a bottle of low percentage alcohol. It is taken by the patient usually on a daily basis.
50, 100 & 150ml are made in amber medical bottles of the respective sizes.
These are first filled with 10% alcohol before adding the medicated granule
10ml % 25ml are filled with 10% alcohol
0ml=st bottle = 10ml bottle with eat dropper
10ml=m bottle = 10ml tincture bottle
25ml bottle = 25ml teat dropper bottle
To Medicate:
The filter paper should be on a clean folded paper towel
Crease the filter paper so that the granule rests in the fold
Place 1 only unmedicated "poppy seed" granule on a fresh 7cm filter paper
Place a drop of the required LM potency onto the granule & leave for 30 seconds to allow excess alcohol to absorb into the filter paper
Carefully lift filter paper & pour the granule into the bottle of 10% alcohol.
Only one granule goes into the bottle regardless of the bottle size
Check to see that the granule has gone into the bottle
You should be able to see it lying on the bottom as it does not dissolve straight away
Put the top securely on the bottle & then apply the label
Instruction Sheet: We have an instruction sheet which gives a standard instruction using a 5ml dose once daily.
Sometimes instructions are required as indicated by the invoice
If the prescriber wants a different instruction we have a similar instruction form where the required dosage can be filled in by the medicator according to the prescriber's requirements.
Put the instruction sheet with medicated bottle either bound to the bottle with a rubber band or folded & inserted with the invoice
LM stock bottle - dispensing.mmp - 01/09/25 - Dr David Lilley - homprac@mweb.co.za

B.4 MTS - Helios (UK) Manufacturing Document

Helios Homoeopathy Ltd.
97 Camden Road
Tunbridge Wells
Kent TN1 2QR
United Kingdom

LM Procedures

LM Overview

The LM dilution scale is one in which each dilution step reduces the concentration of dissolved substance to 1/50,000th of the preceding concentration. We could equally state that 1 drop is added to 49,999 drops.

The other factor which differentiates LMs from centesimal and decimal potentising is that each dilution step is followed by 100 succussions rather than 20.

The above 2 statements describe the basic action of LM potentising. The details of how this is done are outlined in the following paragraphs.

Please note also that LMs are done in 96% alcohol rather than 90%, and all references to alcohol mean 96%.

Starting the LM scale:
Hahnemann established the starting potency from which the LM scale should start as 3C. That is, 3C is the potency which becomes diluted by 50,000 to create the first LM potency or LM 1. It would seem simple enough to add 1 drop of 3C to 49,999 drops of alcohol and succuss to create the LM 1, but this would mean adding a drop of 3C to approximately 1.7 litres of alcohol and succussing this 100 times. The volume of alcohol needed and the weight and size of the succussion bottle make this unworkable in terms of both cost and effort. The first dilution is therefore done in a sequence of 2 dilution steps as follows:

1. First, the 3C is diluted by 500 – i.e. adding 1 drop of 3C to 500 drops of alcohol (about 17mls) in a 30ml medical bottle. This is not succussed but gently swirled to achieve thorough mixing.
2. One drop of this is then added to a further 99 drops (1 dispensed dose of alcohol = 3.4mls)) in a culture tube and this is then succussed 100 times and labelled as "LM 1" on the cork.

The result of the 2 dilutions is a total dilution of 1:50,000 (1/500 x 1/100 = 1/50,000). Note that the succussion is done after the second dilution so that a full 1/50,000th dilution occurs before the succussion.

The remainder of the 500-drop bottle is labelled and retained for subsequent preparations of LM 1. The bottle is labelled with the remedy name followed by "LM Ø" as it serves as our starting "tincture" for the LM range of potencies.

LM Granules MEDICATION
These are 2 gram vials of "poppy seed" granules medicated with LM potencies of medicated alcohol. These granules are smaller than those used for the centesimal medication and are kept in the cupboard in the LM area. The granules are used by practitioners to make up LM tinctures for their patients.

On our invoices they are indicated by the size code: 2g.

Take one of the plastic trays that come with the 4 gram, 8 gram, etc. tablet bottles. Wipe it clean with alcohol and place a suitable length of paper towel in it to line the bottom. Even better, two squares of kitchen towel each folded in half and placed side by side is an ideal method of lining the tray because it gives a double layer of towel thickness to minimize contamination from the tray (which will have had previous potencies on it).

Take one 15 cm filter paper and fold it in half twice and then write the name and LM number near the curved edge of the folder paper. Open this into a cone and clothespeg this by its edge onto the ring on the ring stand.
Pour one 2 gram vial-full of "poppy seed" granules into the cone.

Drop 10 - 12 drops of the LM potency onto the granules. Let this stand for a few of minutes to allow the alcohol to flow through the granules. Make sure the alcohol on the granules doesn't also soak upward on the filter paper to the point where it contacts either the clothespeg or metal ring, otherwise there is a danger of contaminating subsequent remedies.

Remove the cone from the ring, collapse the cone (i.e. fold it flat), fold the edge down and then shake the granules around to ensure that all granules are thoroughly medicated.
Place the folded cone flat onto the paper towel in the tray with the remedy name at the edge of the paper exposed to view.

Continue preparing LMs in this fashion, placing them in the tray ensuring that they are well away from each other to prevent cross-contamination. The maximum number of LMs to a tray is 4. Allow these to dry thoroughly. This takes approximately an hour in winter and about a half hour in summer.

Once the LMs are dry they are individually lifted from the trays, and the filter paper partially unfolded and carefully pressed by hand to break up the granules which have now clumped during the drying process. The granules are then poured into 2 gram vials, the plastic tops are put on and they are each labelled.

Appendix C - NMR Analysis Approval

Nuclear Magnetic Resonance
CAF Central Analytical Facilities
STELLENBOSCH UNIVERSITY

3 November 2014

Durban University of Technology:

Mr Garth Miller will have access to perform his NMR work at our NMR unit.
He will partake in the process by preparing the samples himself and overseeing the acquisition of the spectral data on the NMR instrument.

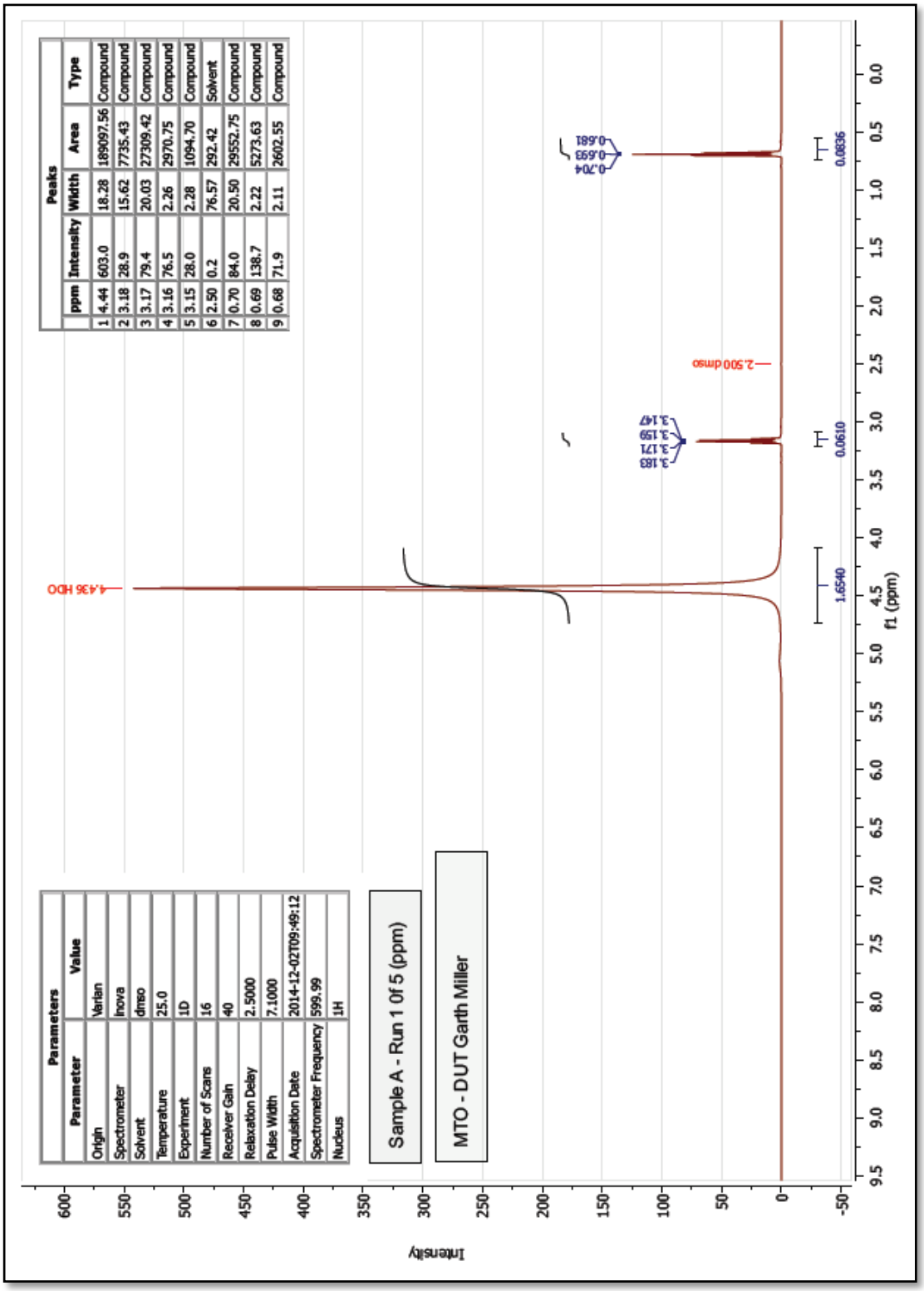
Kind Regards
Dr DJ Brand.
Nuclear Magnetic Resonance Unit
Stellenbosch University

NMR Laboratory
Room 1001 (1014), Mike De Vries Bldg., Private Bag X1, Matieland, 7602, South Africa
Tel: +27 21 808 3338 Fax: +27 21 808 3360 djbrand@sun.ac.za http://academic.sun.ac.za/saf/services/services5.html

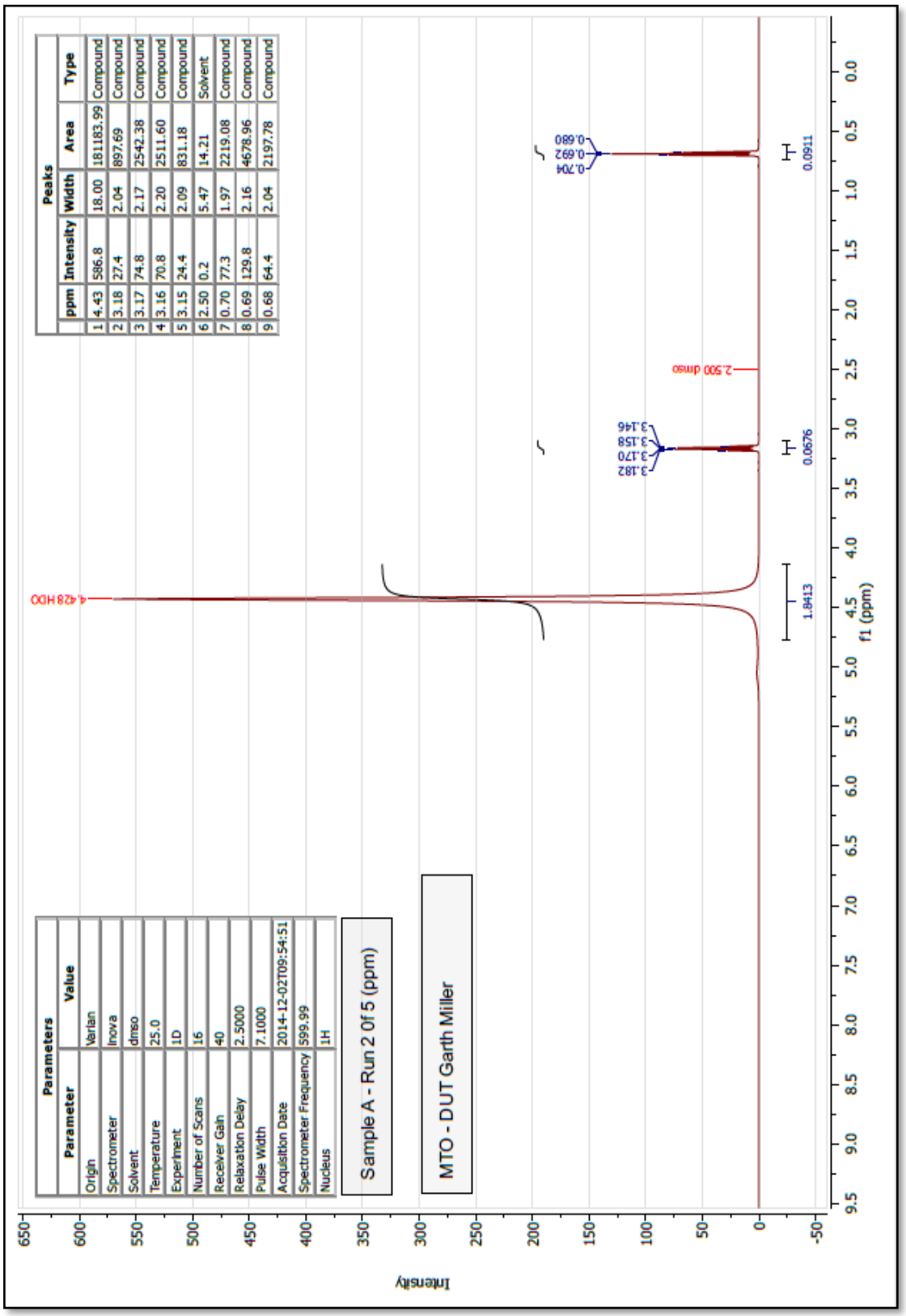
Appendix D - LM1 sample for analysis

- Measure out 1 granule into 250ml borosilicate glass bottle
- Add 118.294ml 20% SVR alcohol solution
- 8 tablespoons at 14.7867ml
- In accordance with Aphorism 248 of the 6th Edition *Organon* (Hahnemann and O'Reilly 2001)
- Allow granule to dissolve completely
- Cap, seal and succuss 10x
    - As would be appropriate for a person of average sensitivity (Hahnemann and O'Reilly 2001)
- Label for detail source
- Randomise labelling for all samples as deemed appropriate (as 'A'-'J')
- Technician to establish a record of random assignments into a sealed envelope.
- Researcher to match samples after analysis.

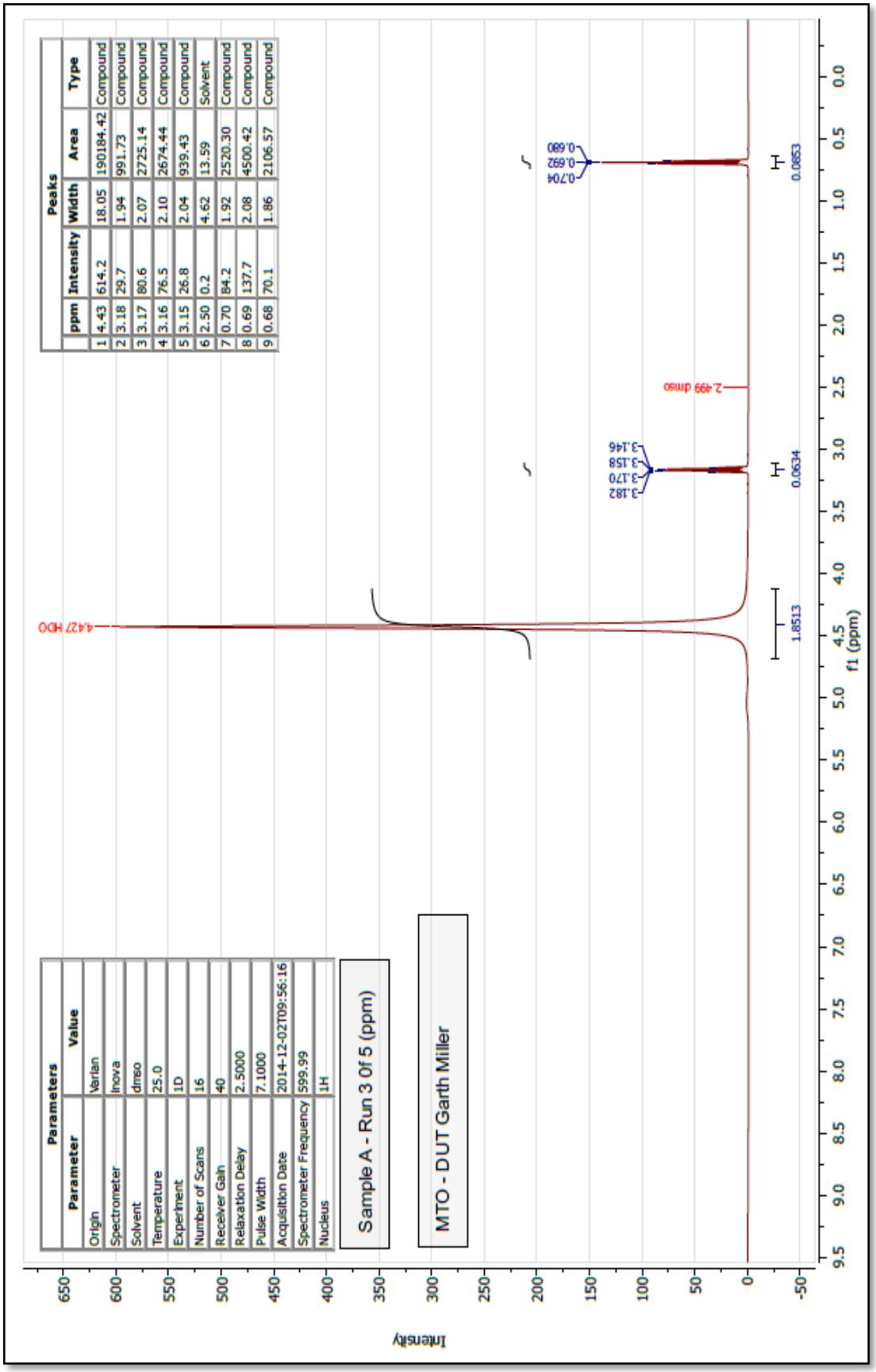
Appendix E - NMR Spectra



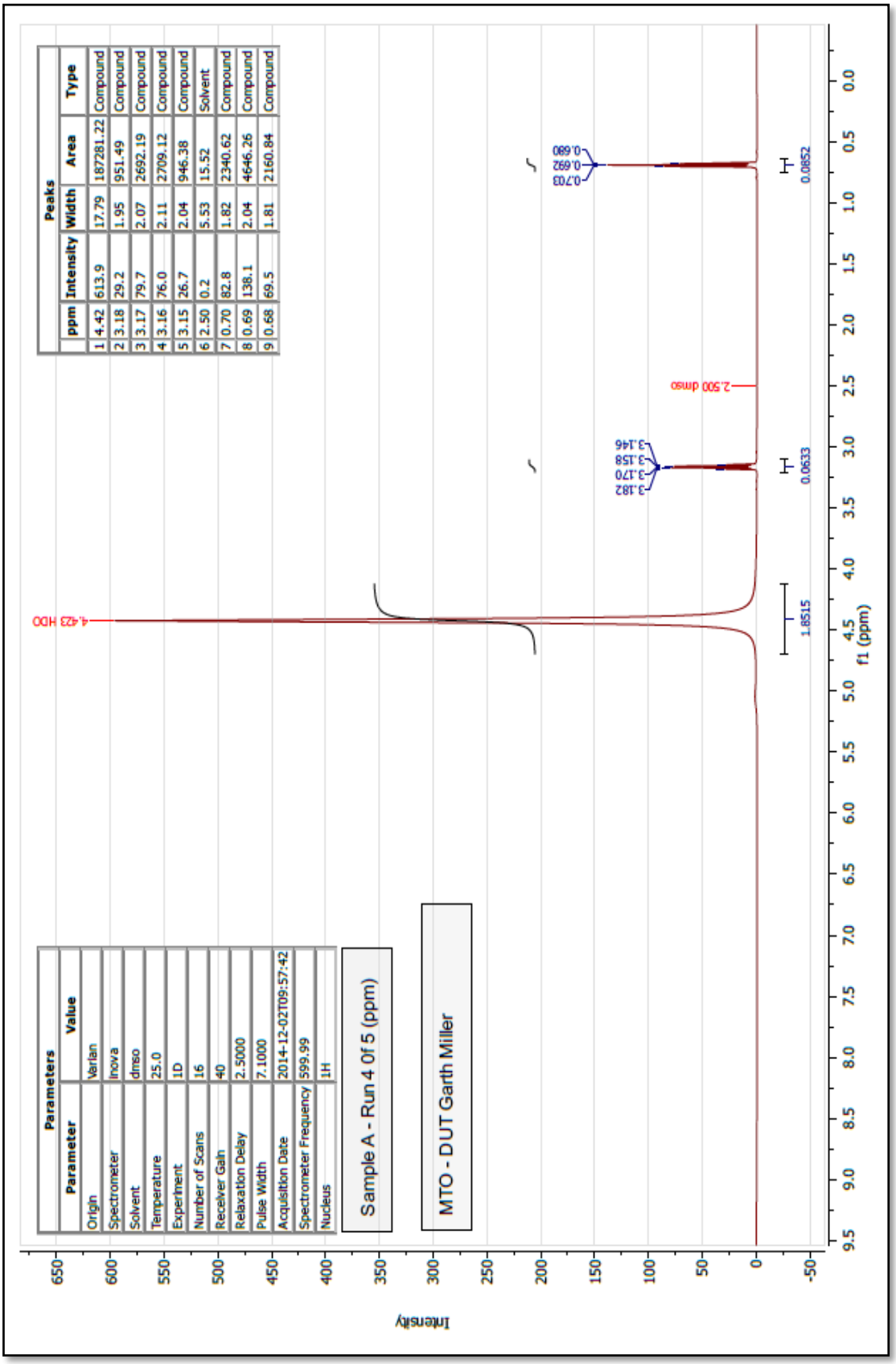
Graph 1: Sample A (Run 1 of 5) MTO - DUT Garth Miller



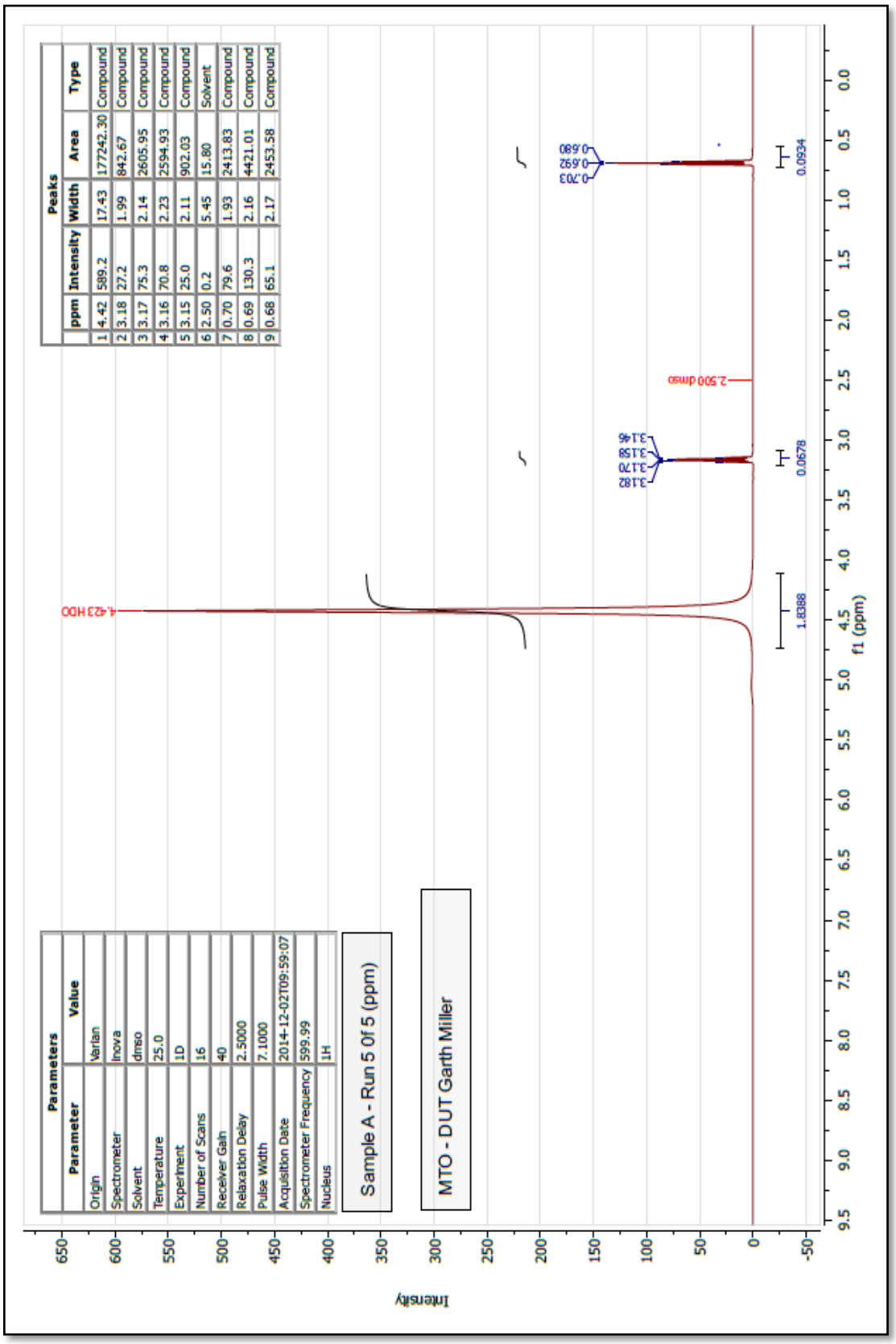
Graph 2: Sample A (Run 2 of 5) MTO - DUT Garth Miller



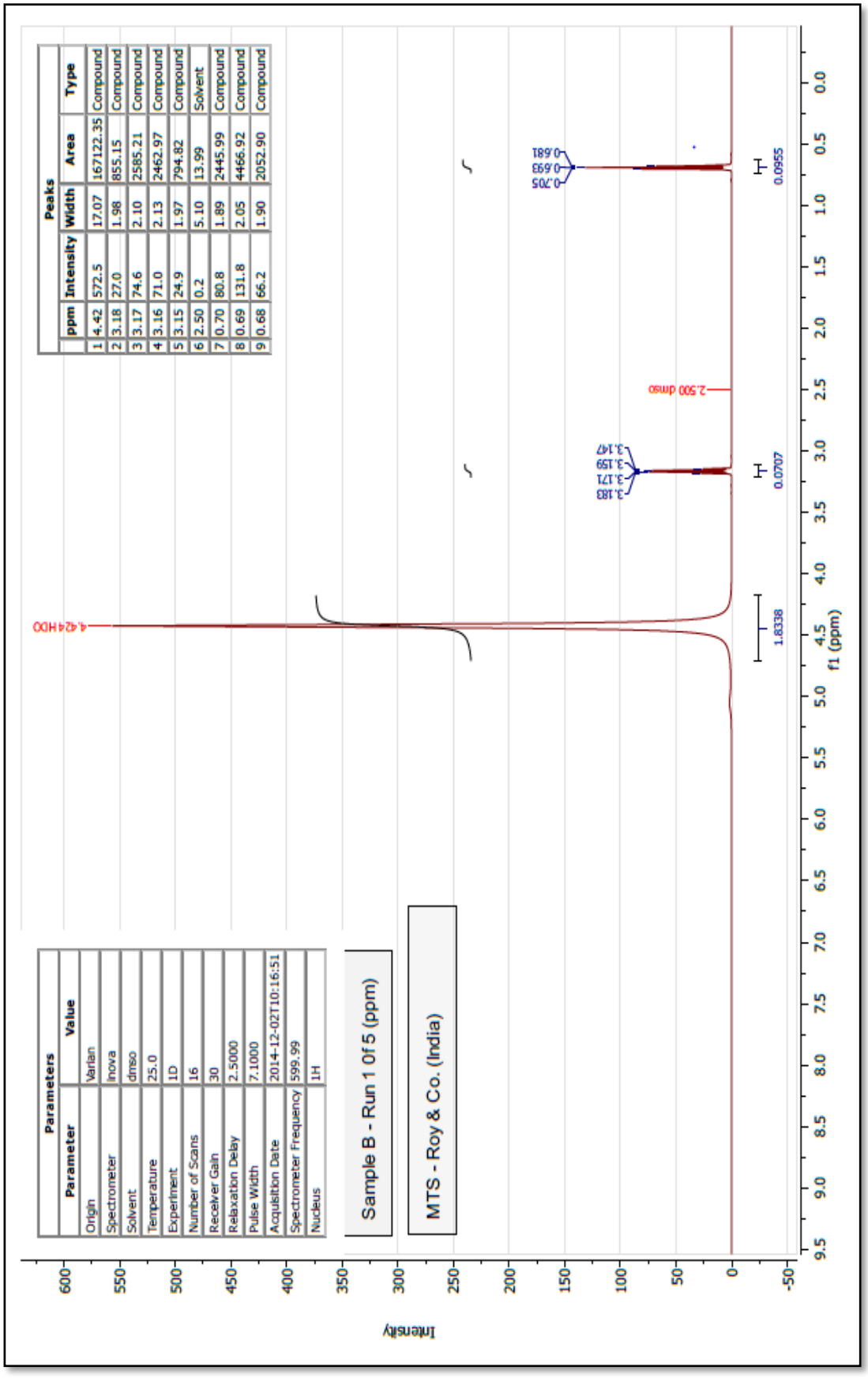
Graph 3: Sample A (Run 3 of 5) MTO - DUT Garth Miller



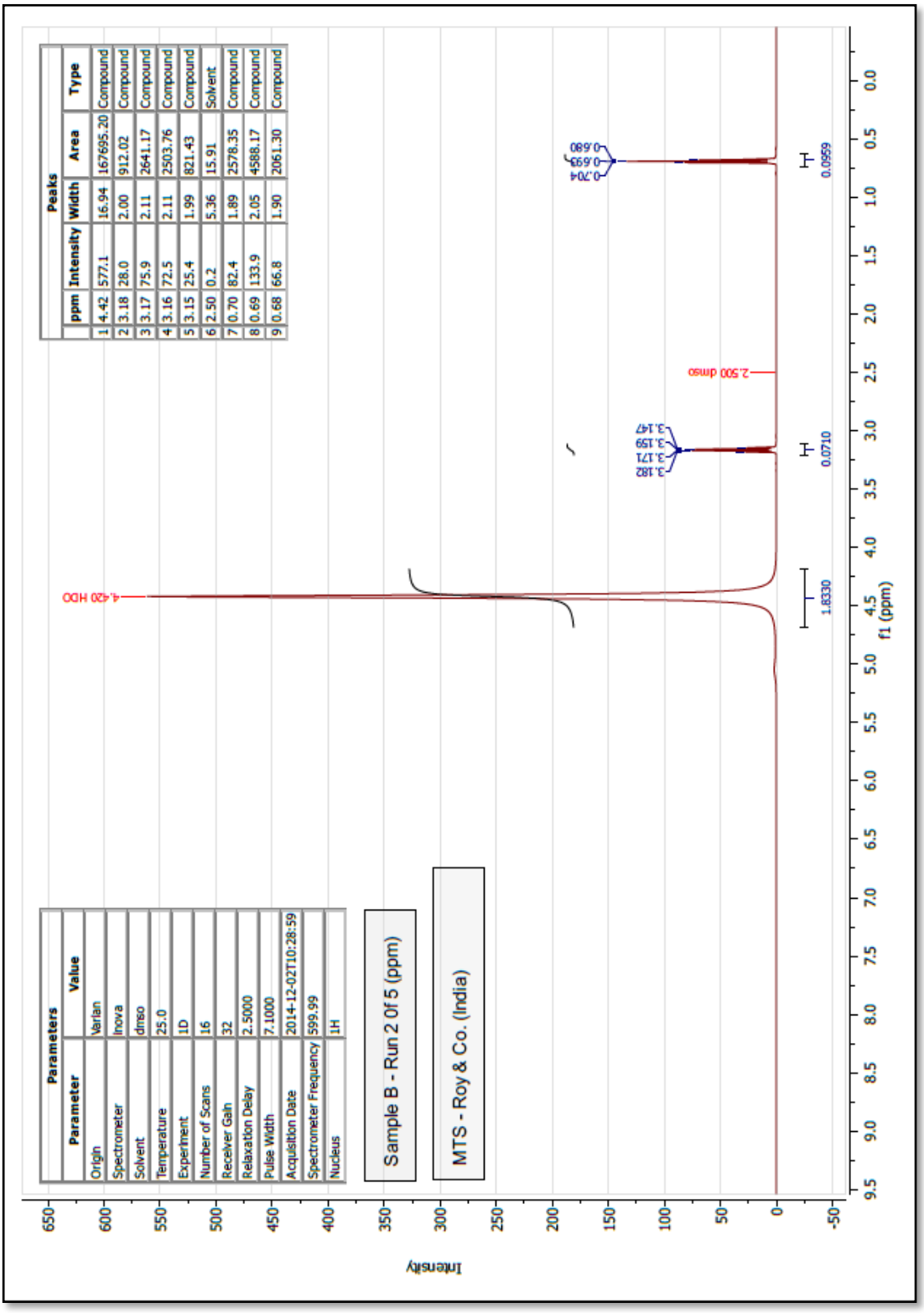
Graph 4: Sample A (Run 4 of 5) MTO - DUT Garth Miller



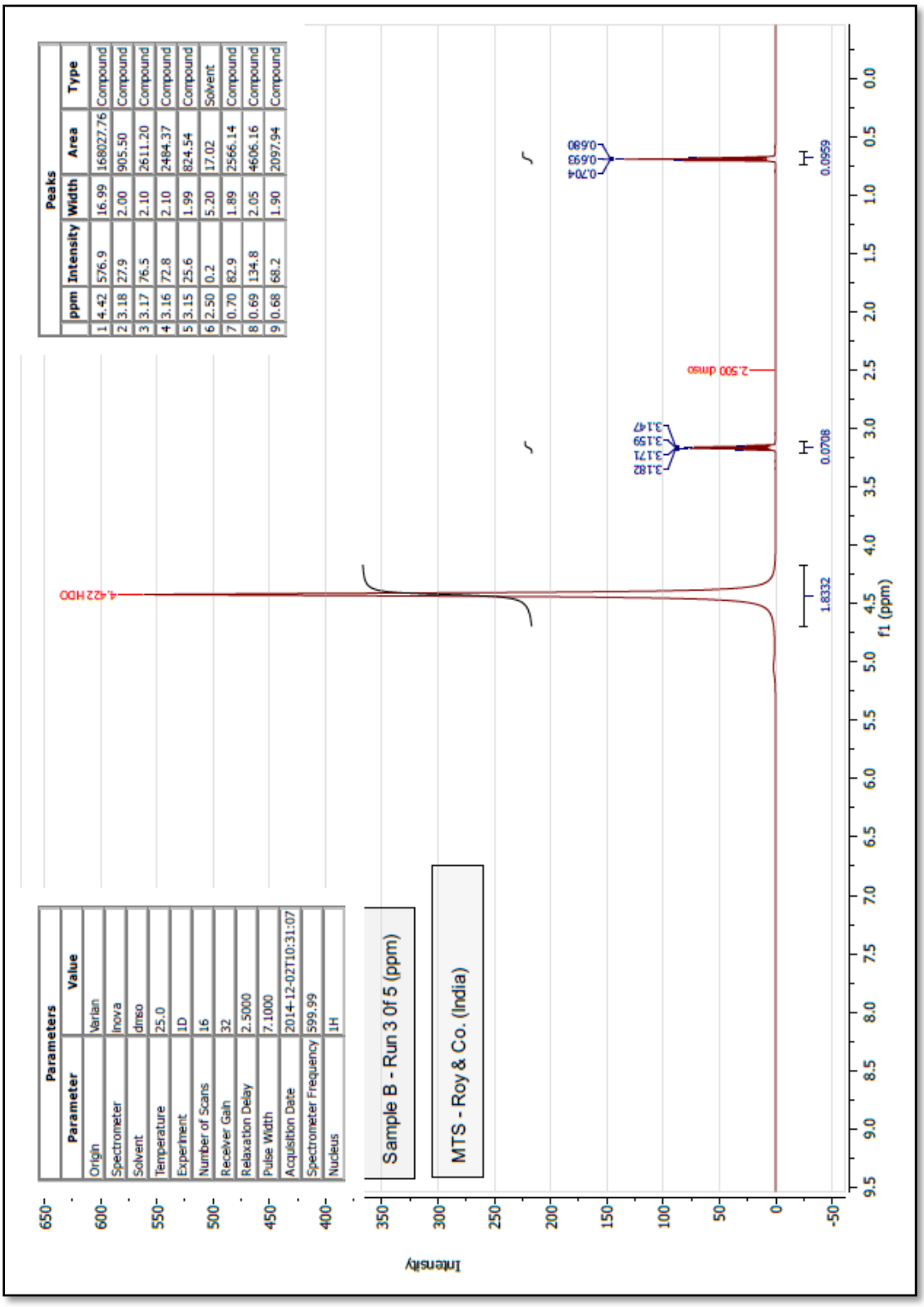
Graph 5: Sample A (Run 5 of 5) MTO - DUT Garth Miller



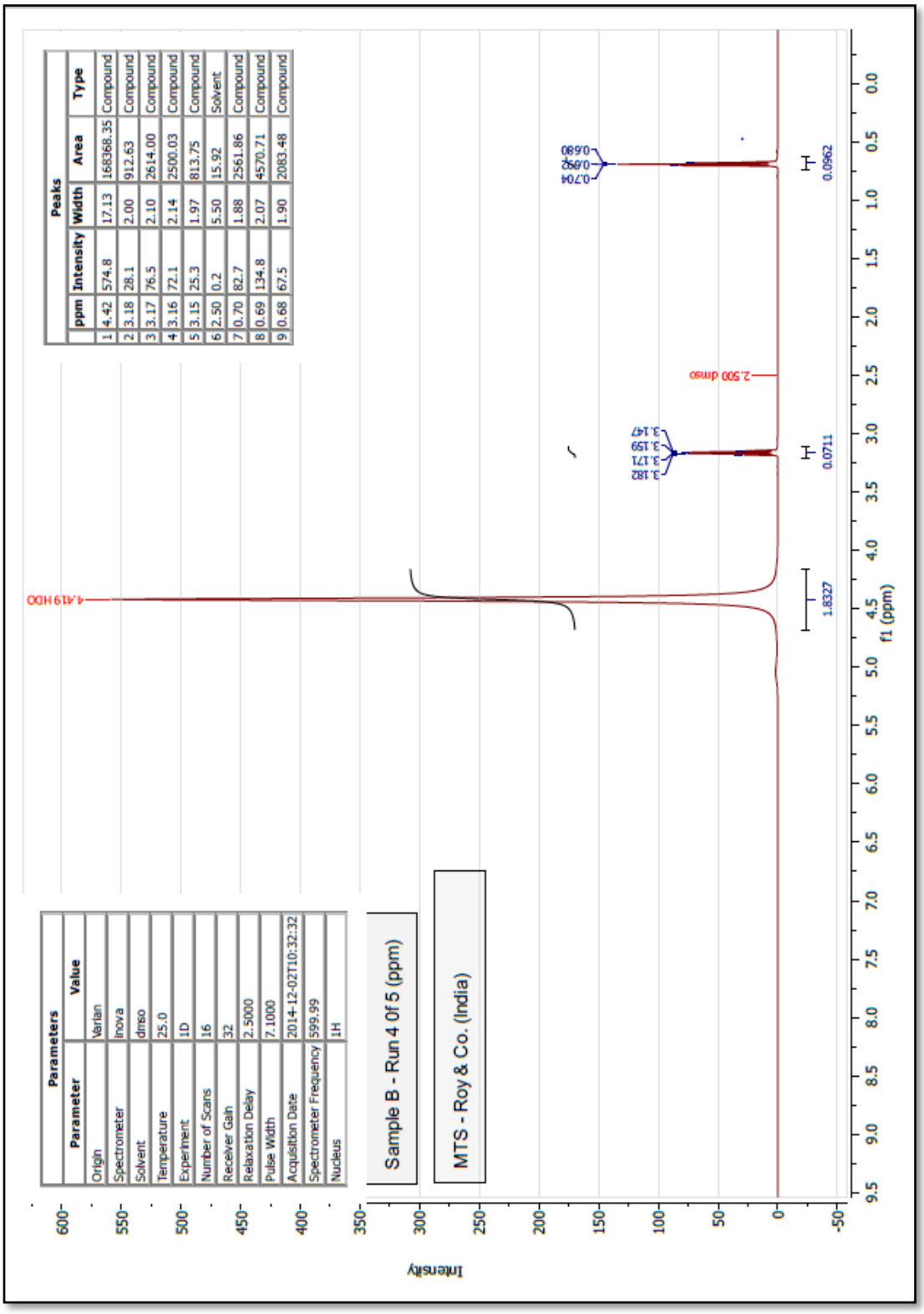
Graph 6: Sample B (Run 1 of 5) MTS - Roy & Co.(India)



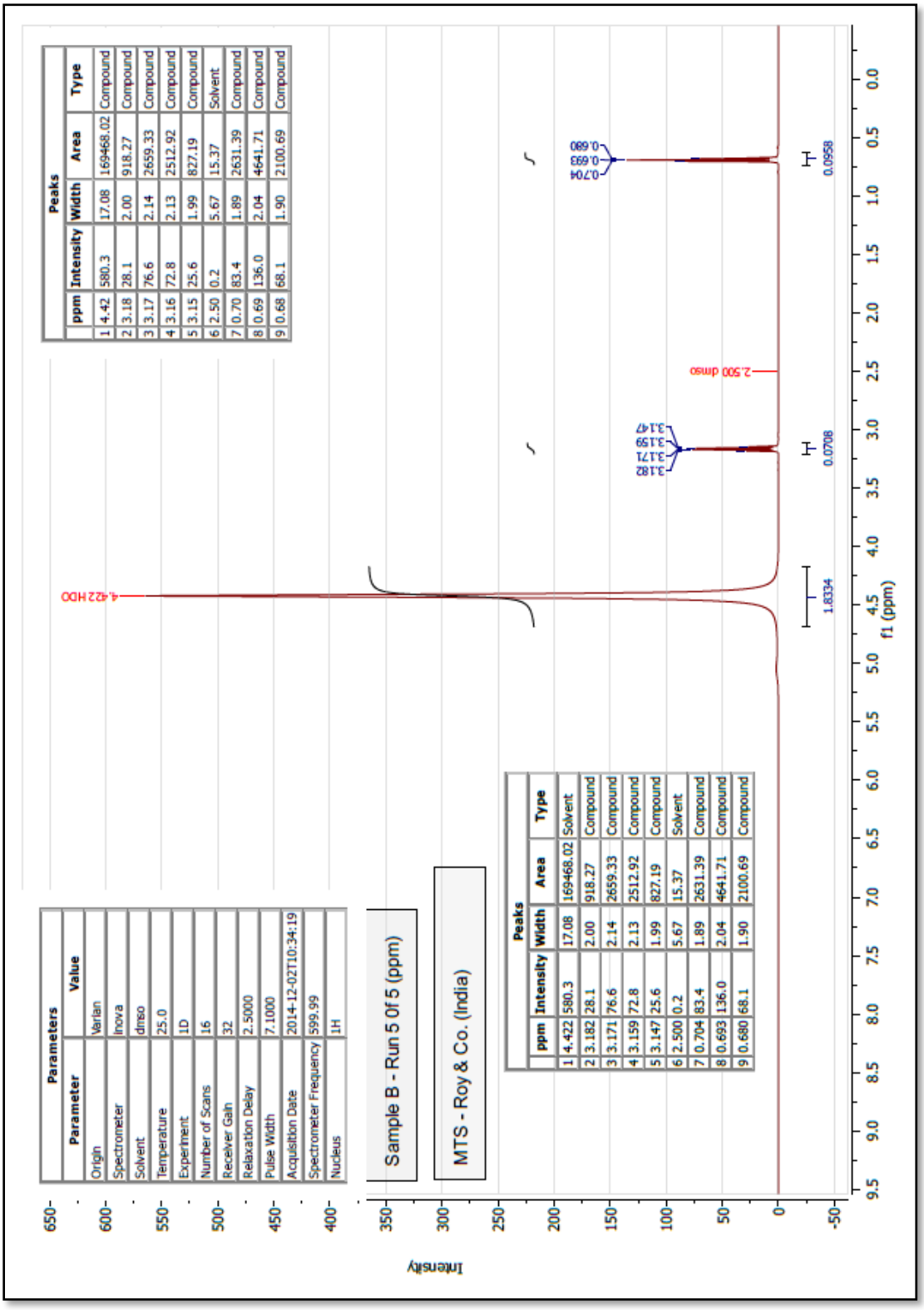
Graph 7: Sample B (Run 2 of 5) MTS - Roy & Co.(India)



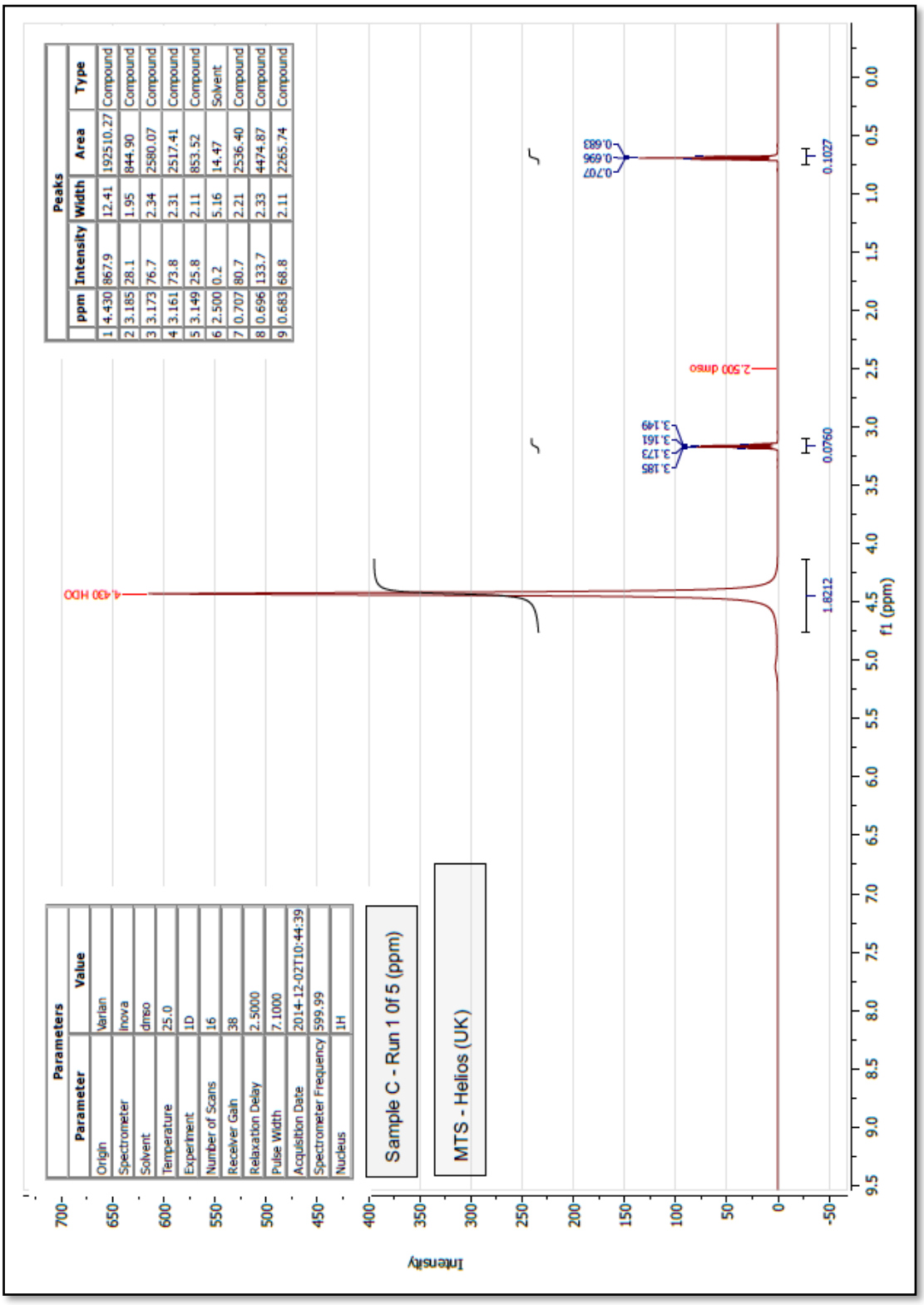
Graph 8: Sample B (Run 3 of 5) MTS - Roy & Co.(India)



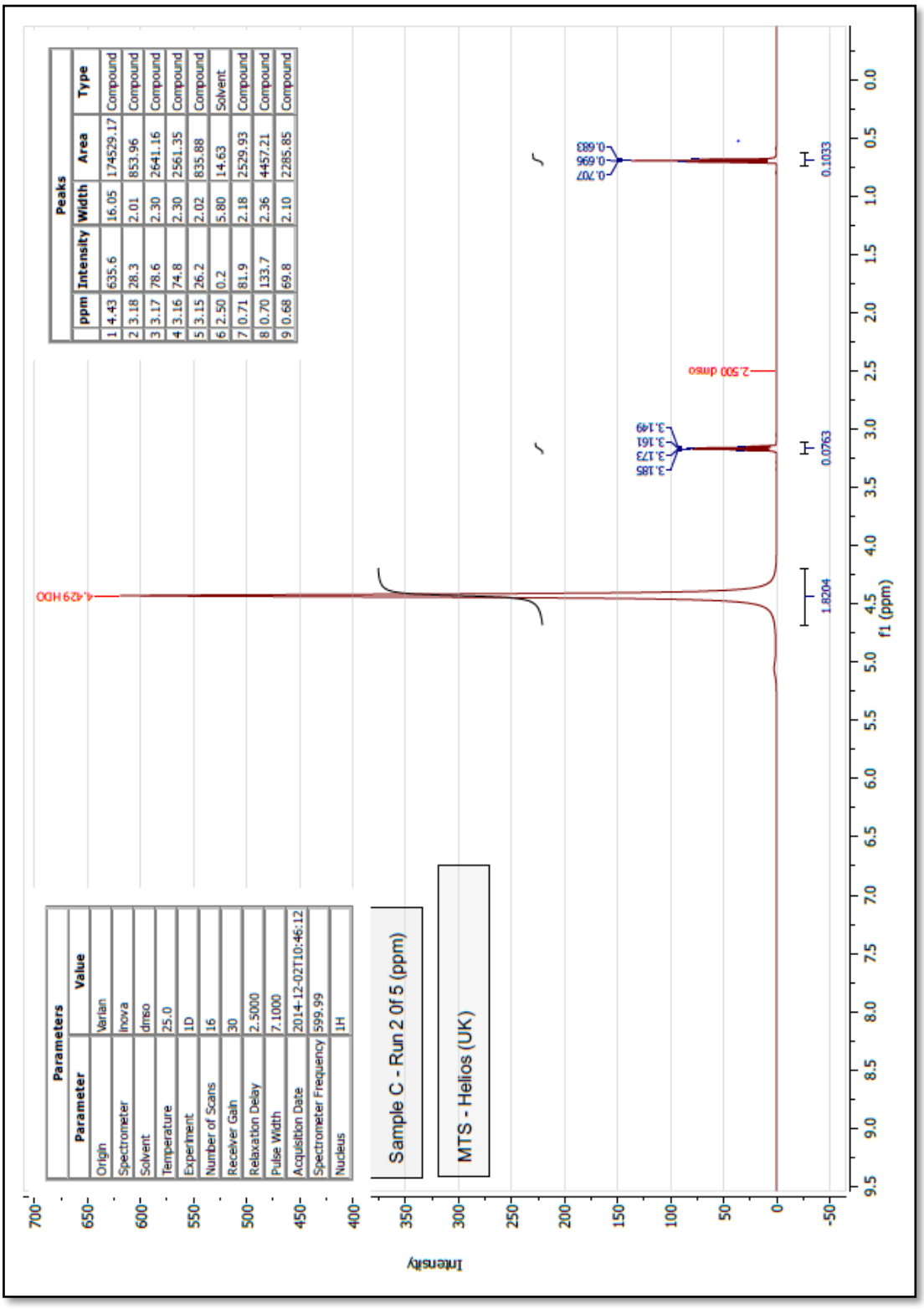
Graph 9: Sample B (Run 4 of 5) MTS - Roy & Co.(India)



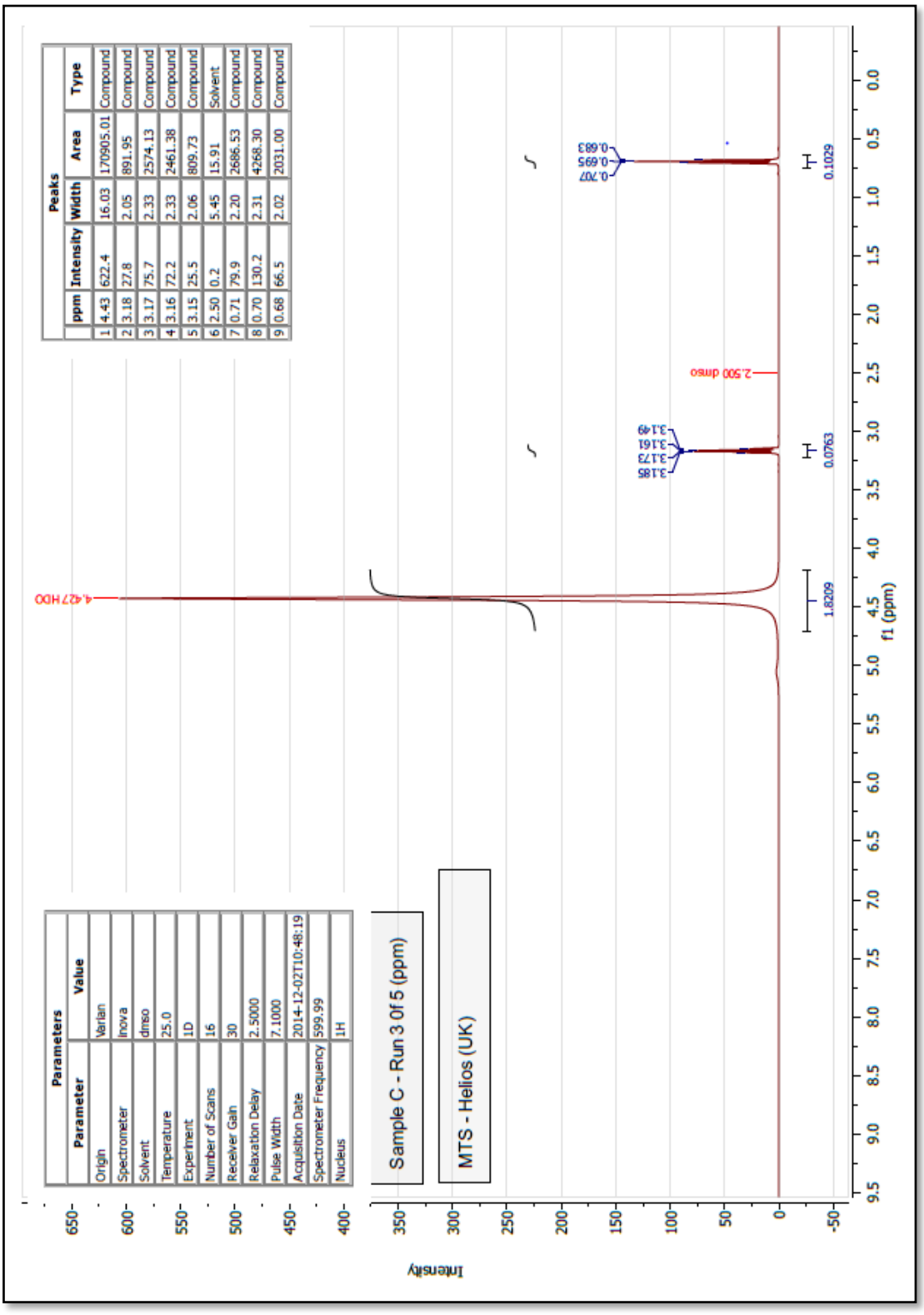
Graph 10: Sample B (Run 5 of 5) MTS - Roy & Co.(India)



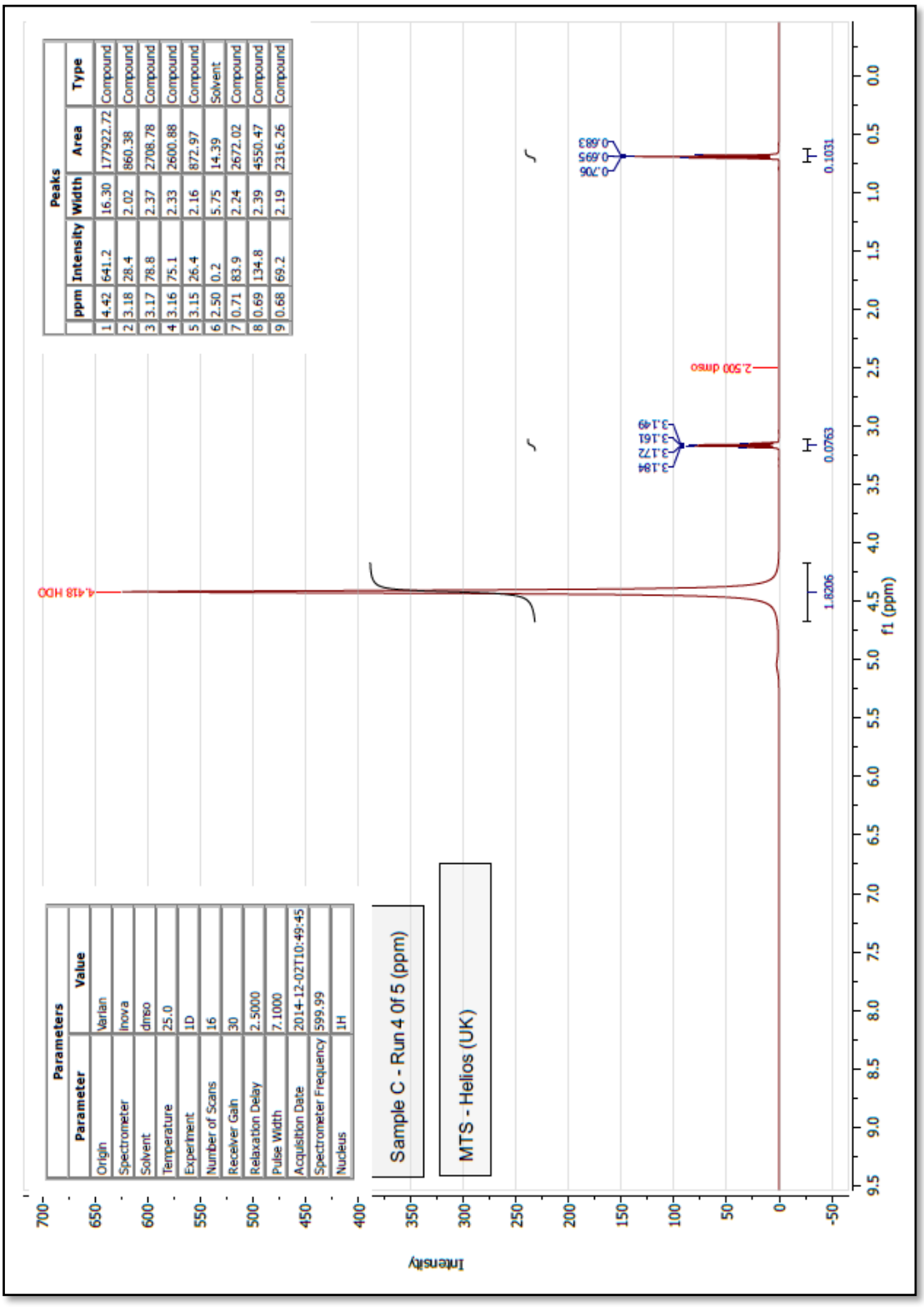
Graph 11: Sample C (Run 1 of 5) MTS - Helios (UK)



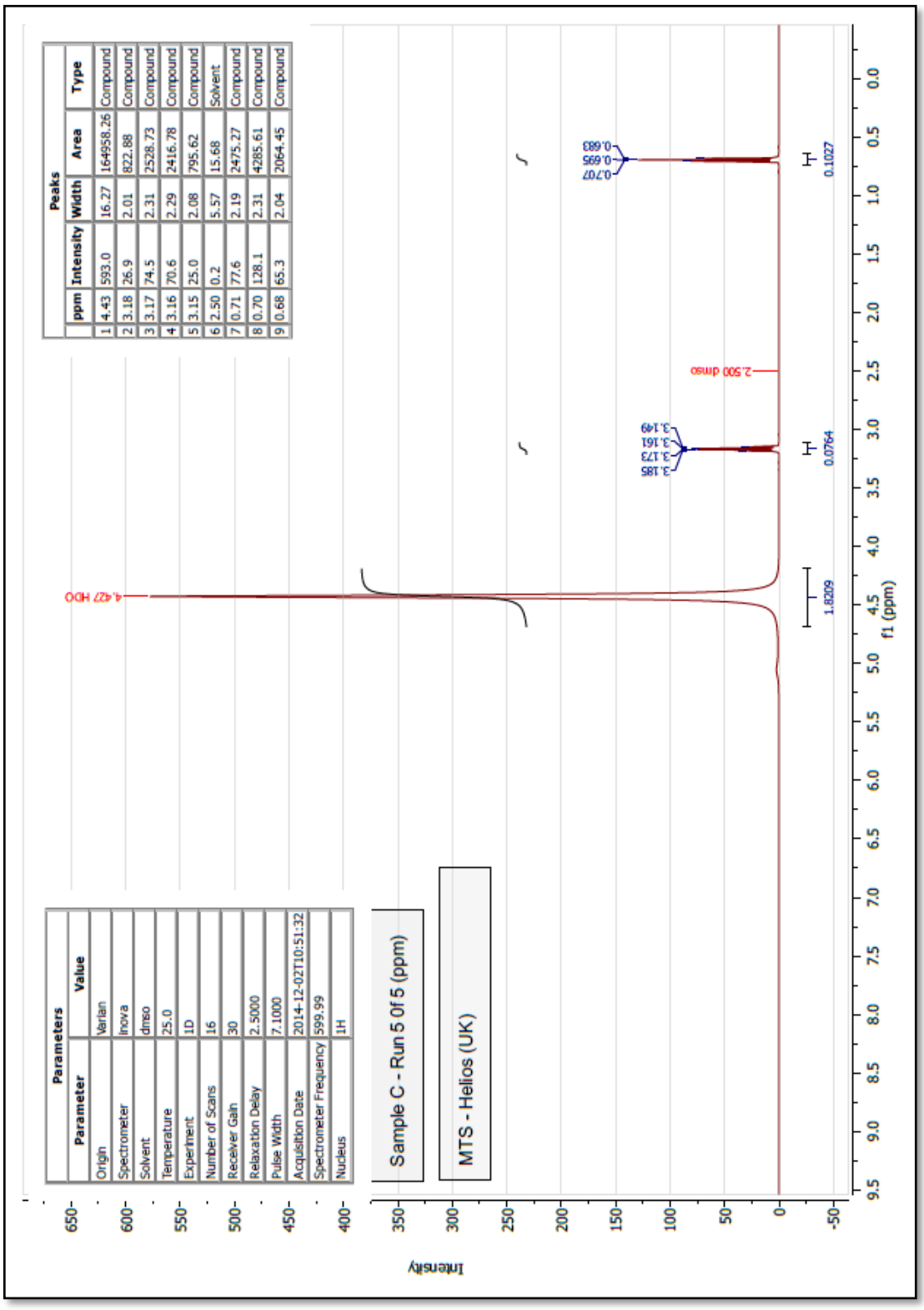
Graph 12: Sample C (Run 2 of 5) MTS - Helios (UK)



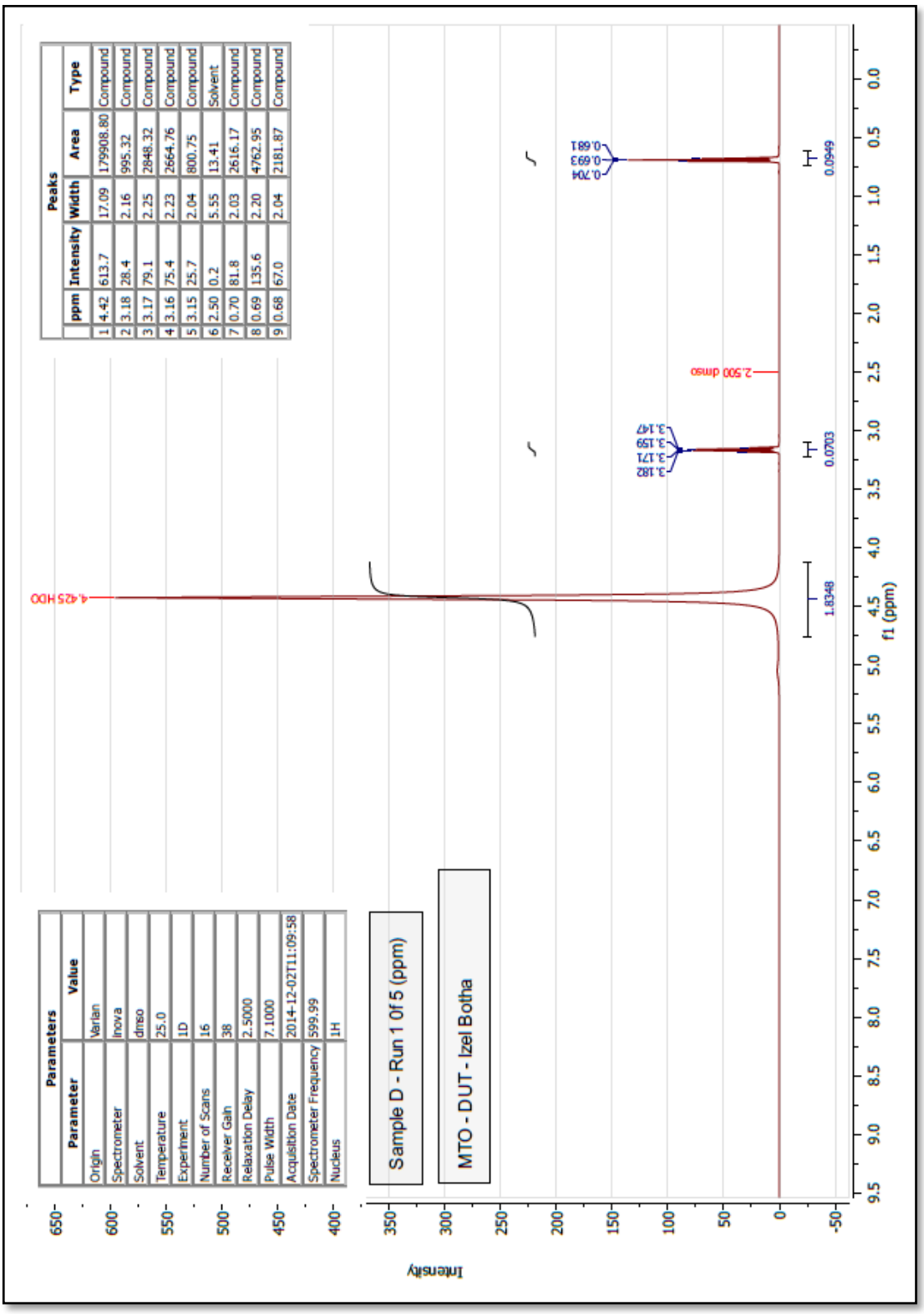
Graph 13: Sample C (Run 3 of 5) MTS - Helios (UK)



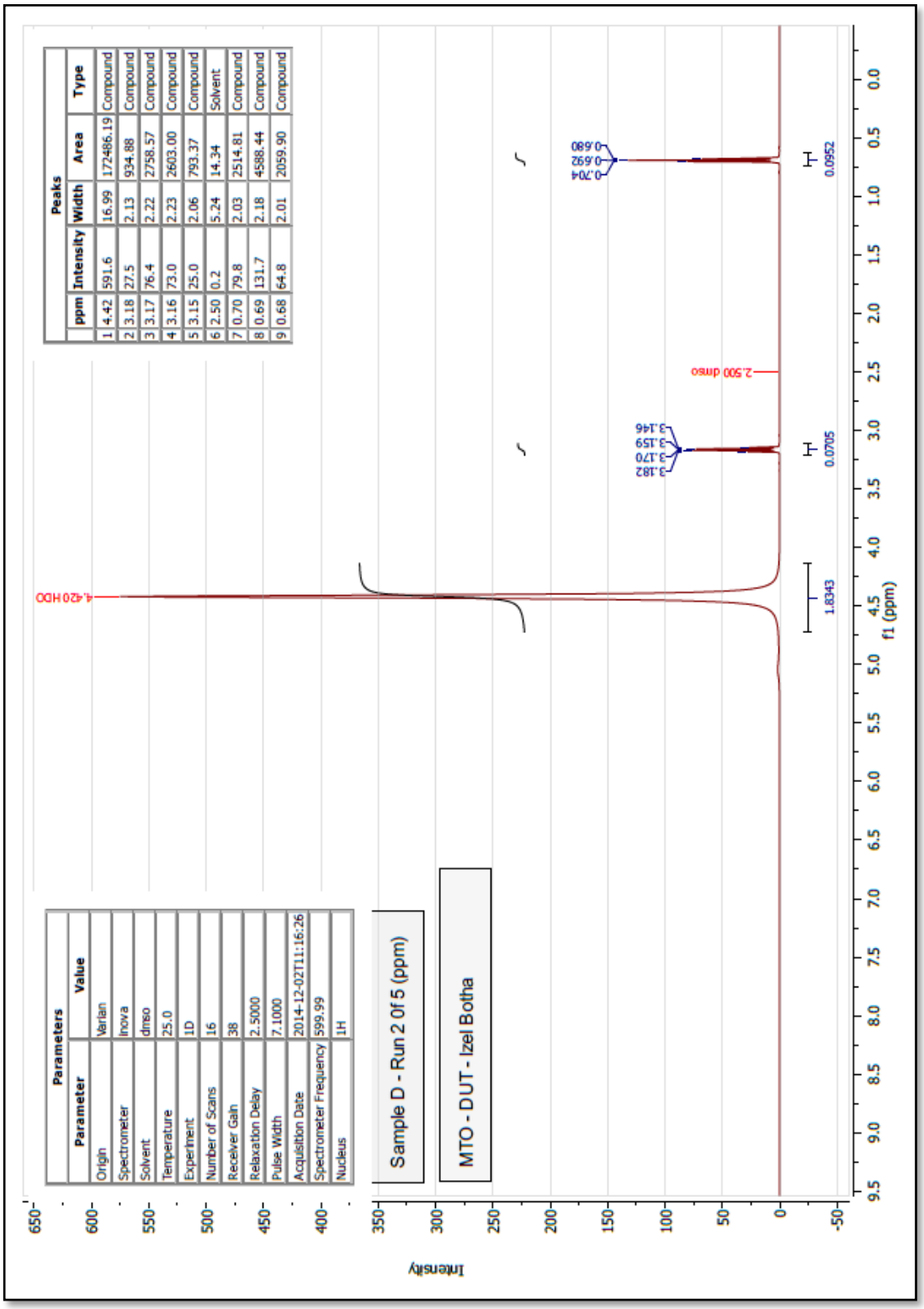
Graph 14: Sample C (Run 4 of 5) MTS - Helios (UK)



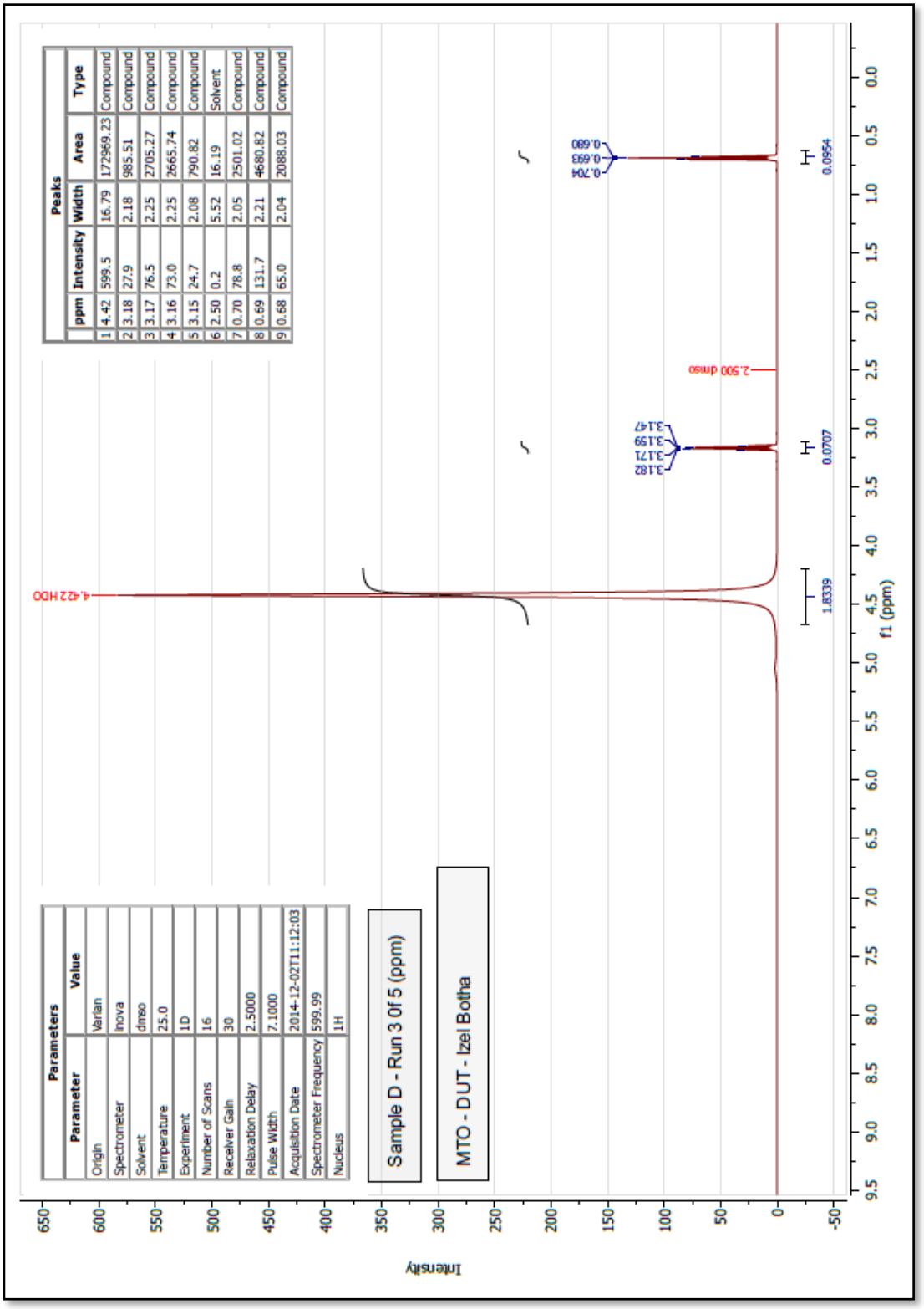
Graph 15: Sample C (Run 5 of 5) MTS - Helios (UK)



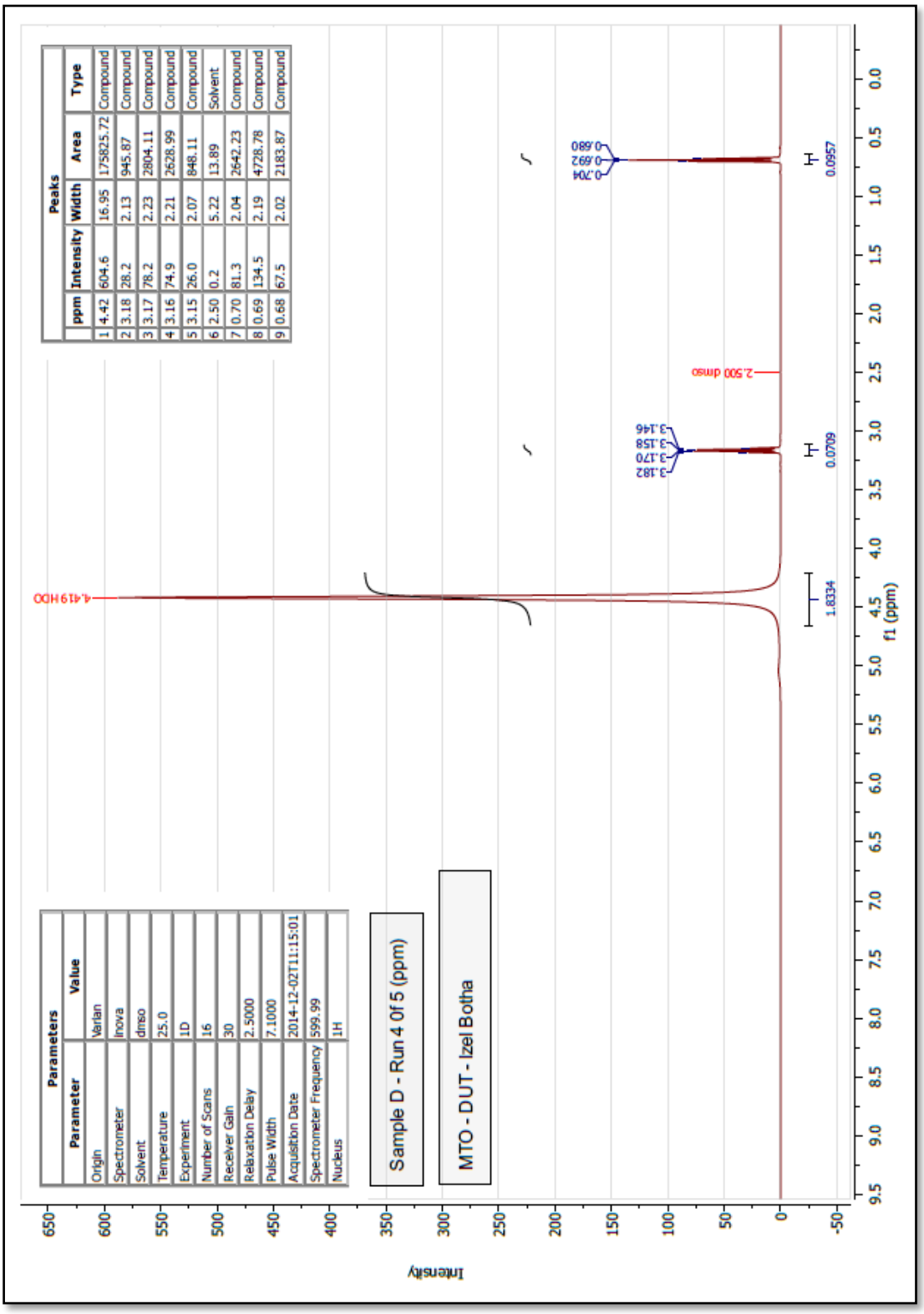
Graph 16: Sample D (Run 1 of 5) MTO - Izel Botha



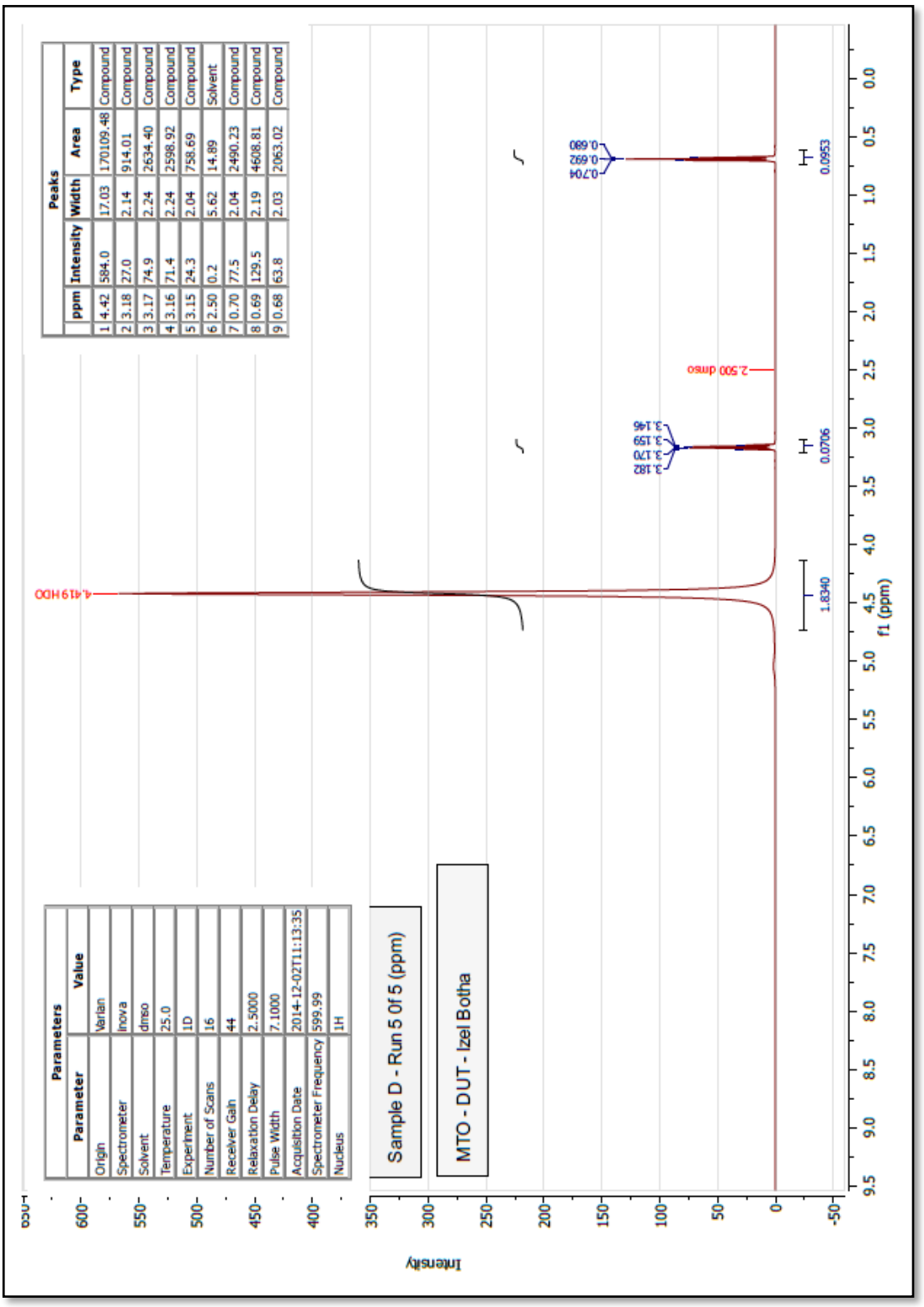
Graph 17: Sample D (Run 2 of 5) MTO - Izel Botha



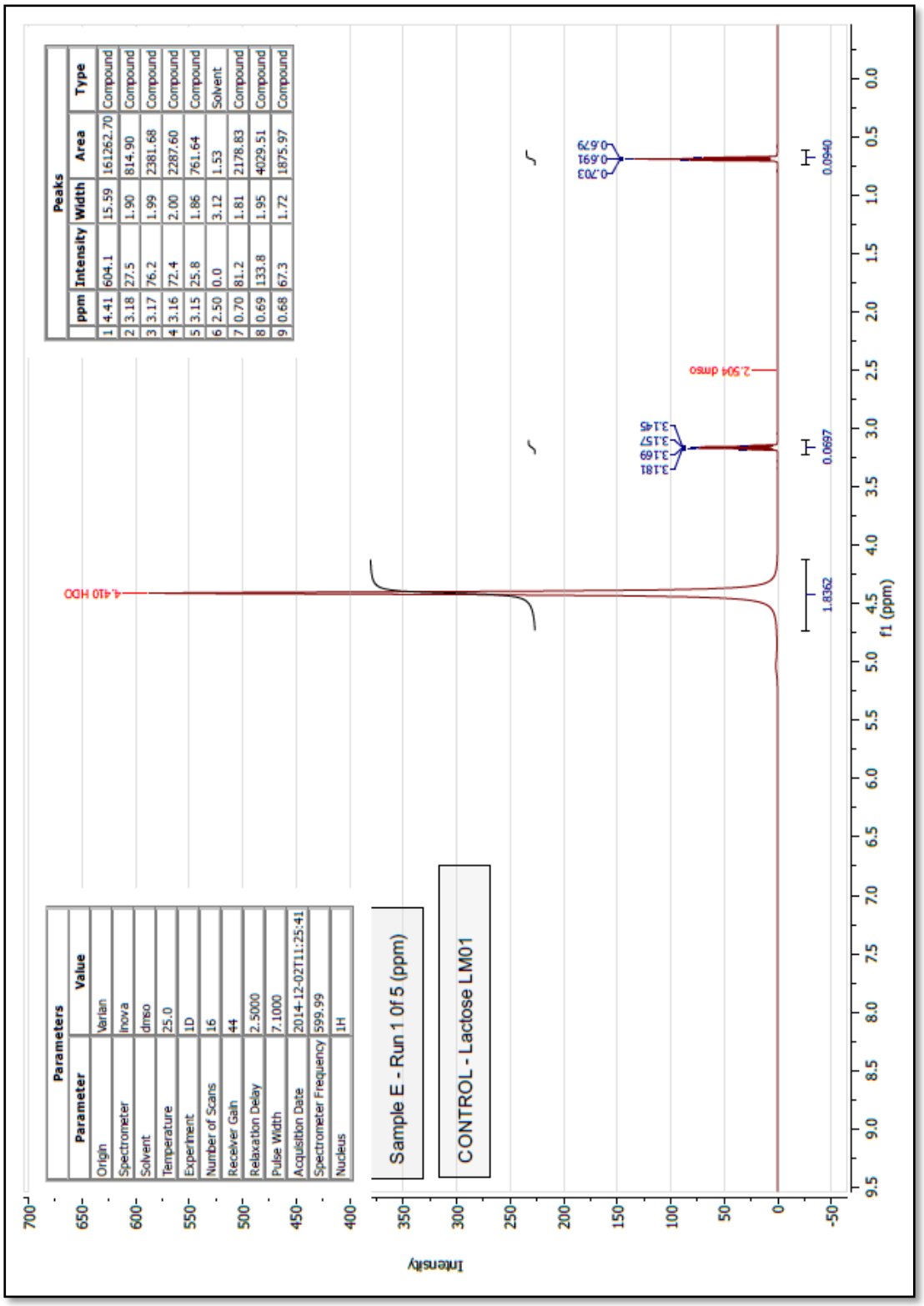
Graph 18: Sample D (Run 3 of 5) MTO - Izel Botha



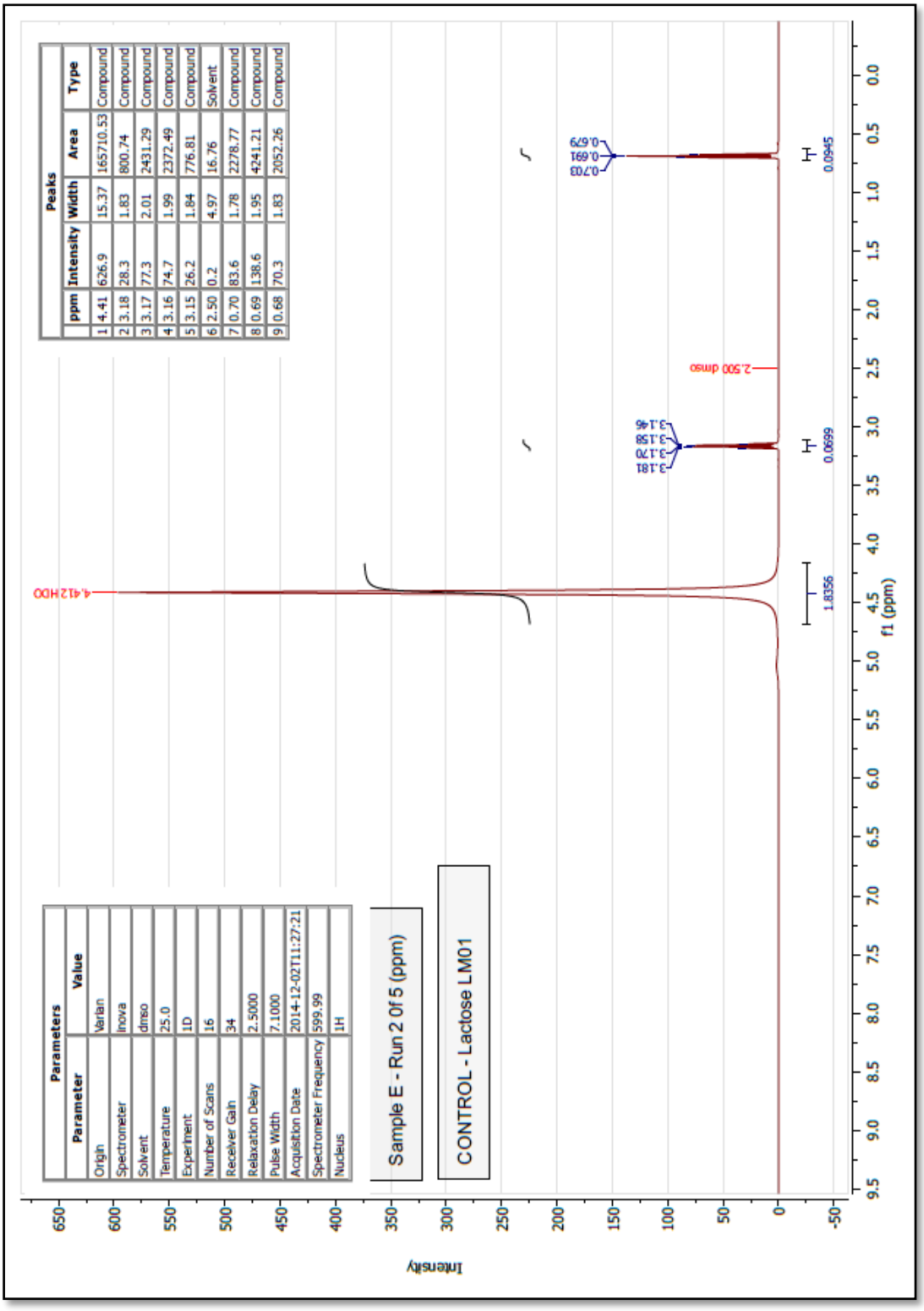
Graph 19: Sample D (Run 4 of 5) MTO - Izel Botha



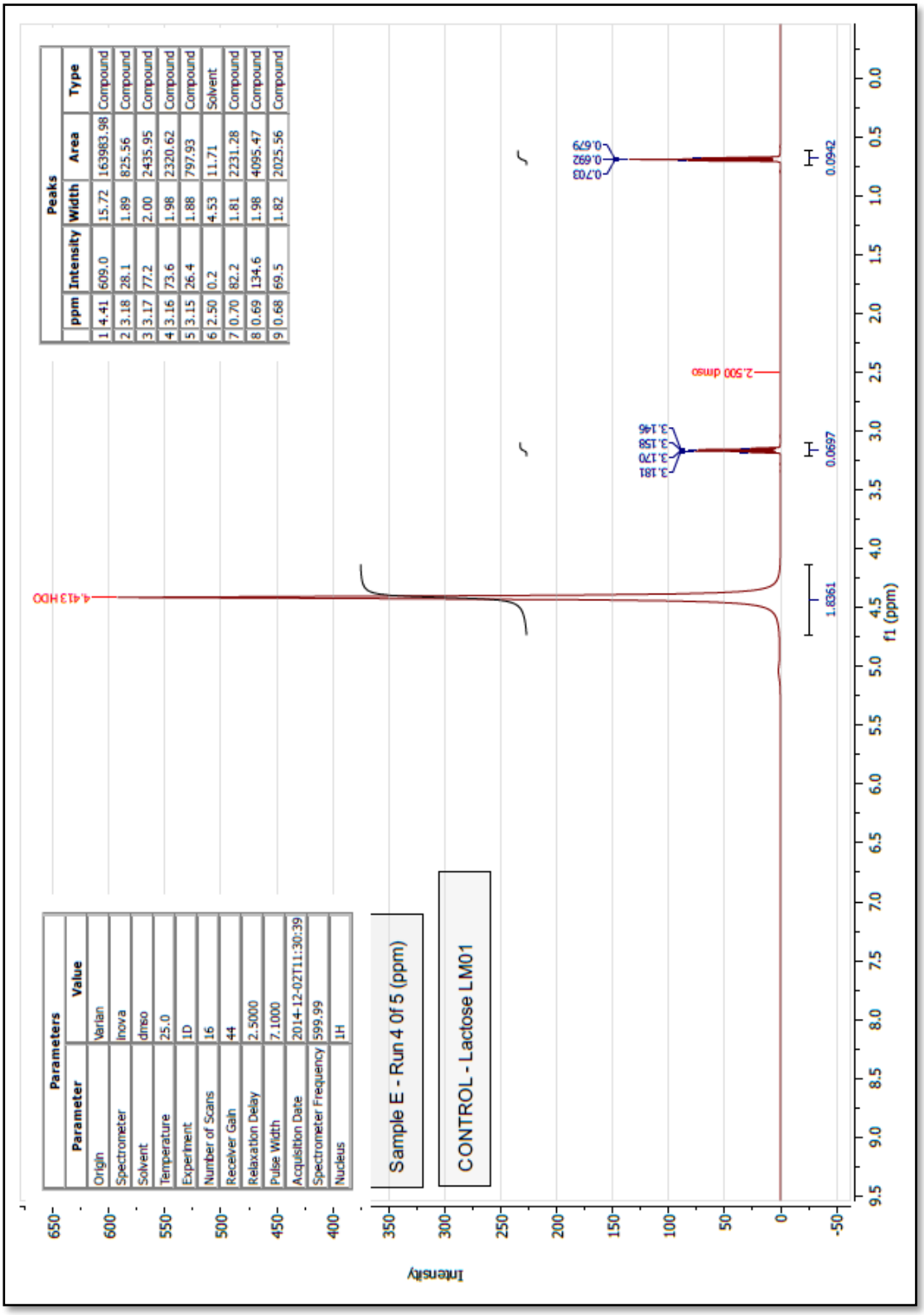
Graph 20: Sample D (Run 5 of 5) MTO - Izel Botha



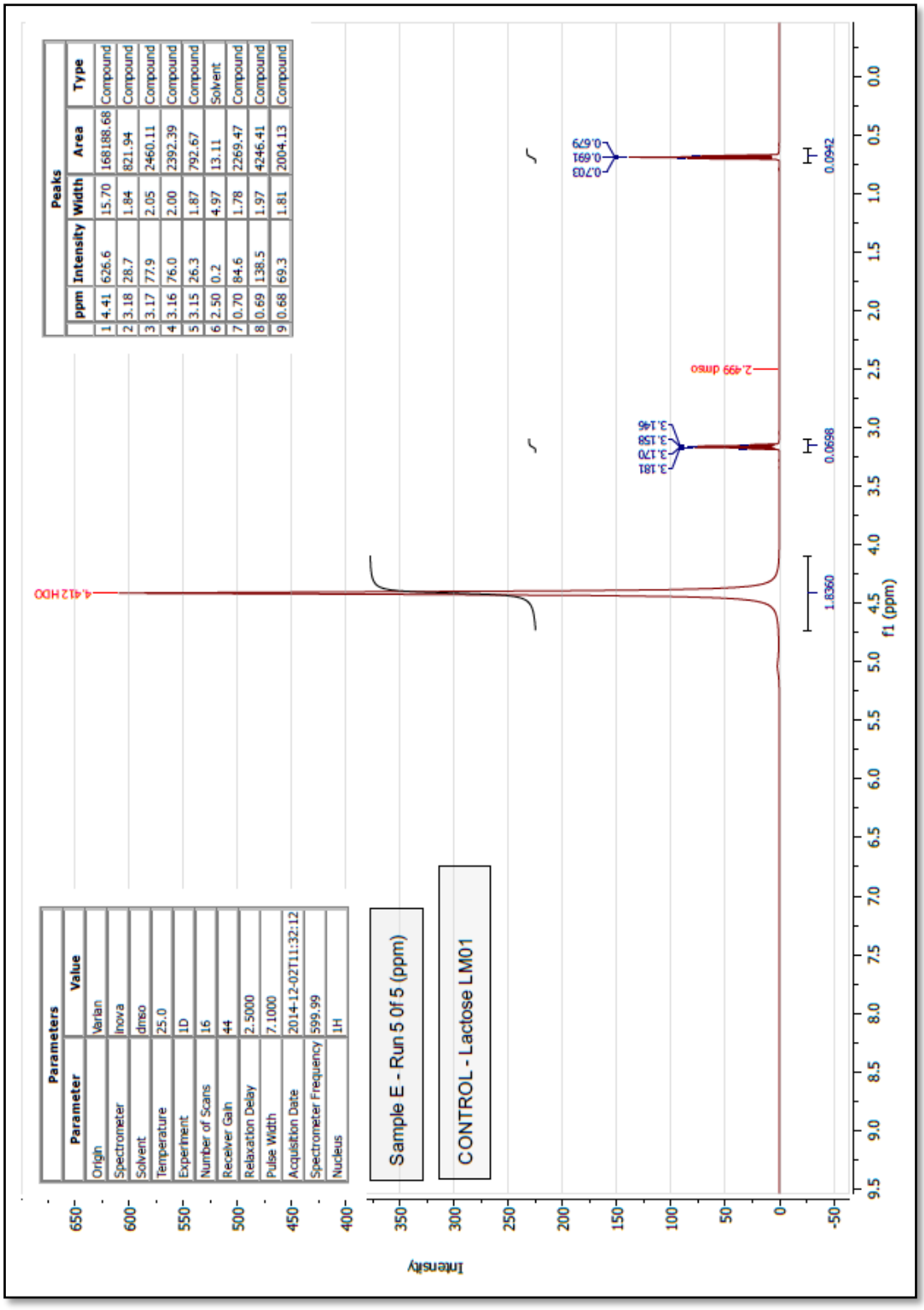
Graph 21: Sample E (Run 1 of 5) CONTROL - Lactose LM1



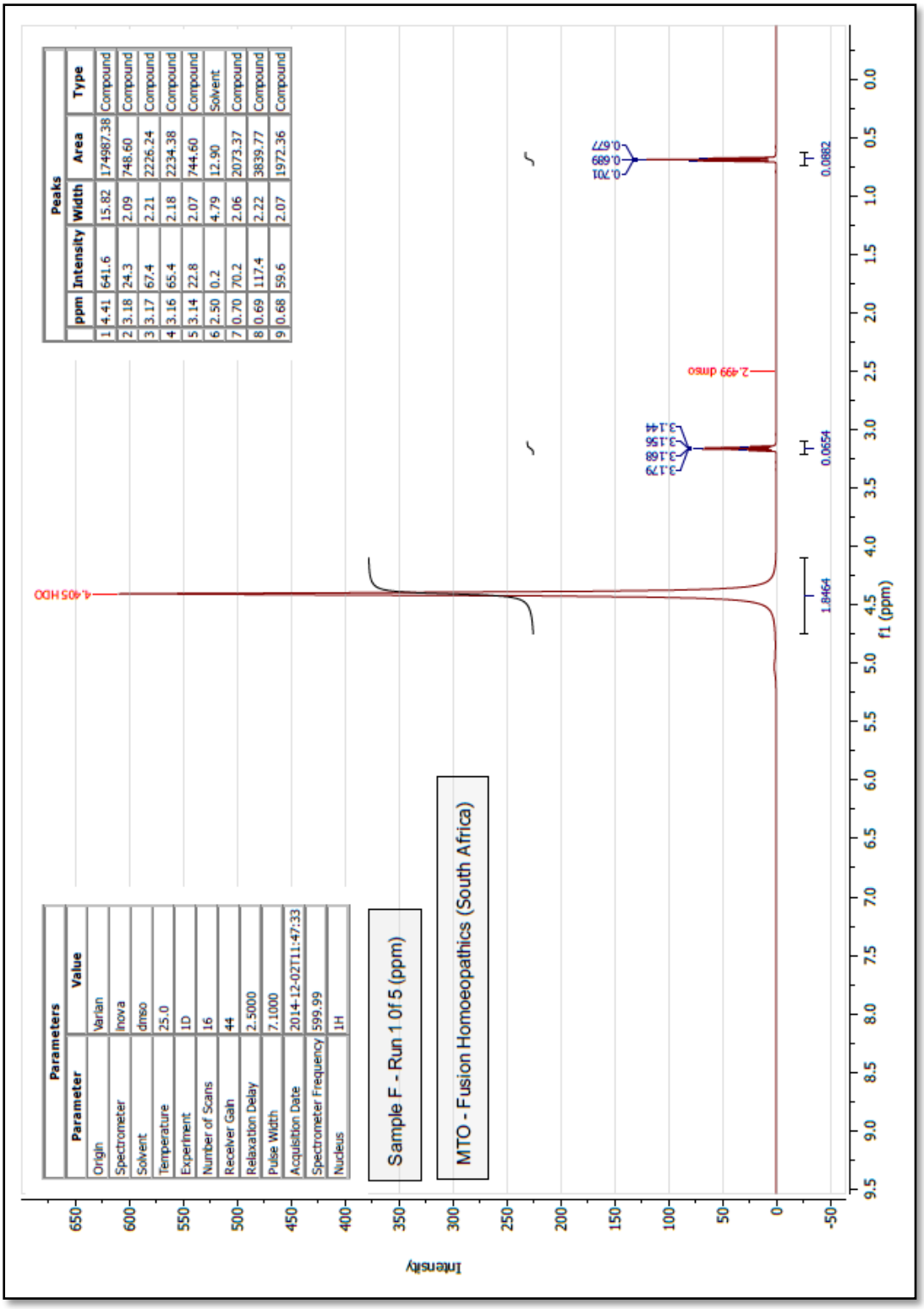
Graph 22: Sample E (Run 2 of 5) CONTROL - Lactose LM1



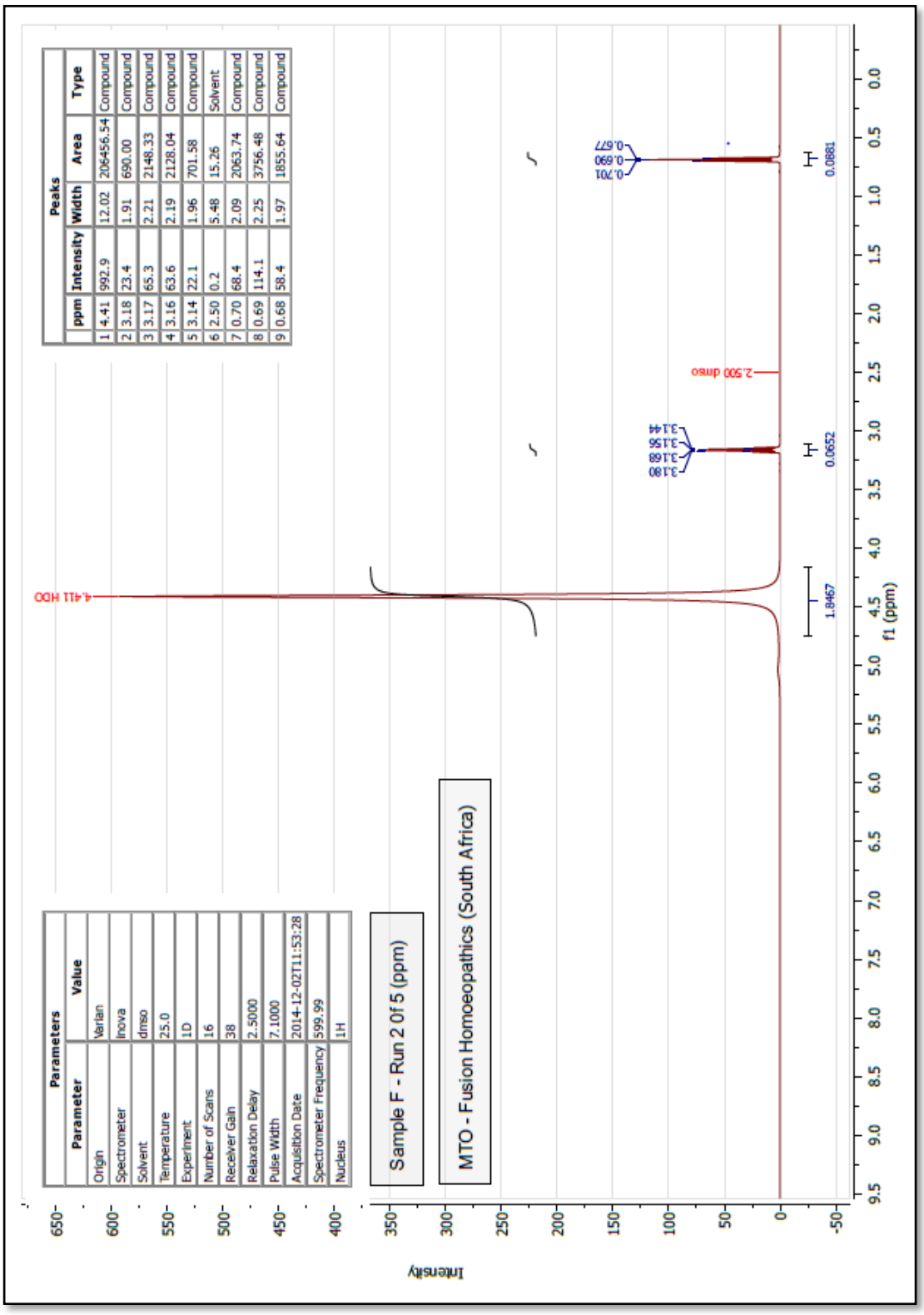
Graph 23: Sample E (Run 4 of 5) CONTROL - Lactose LM1



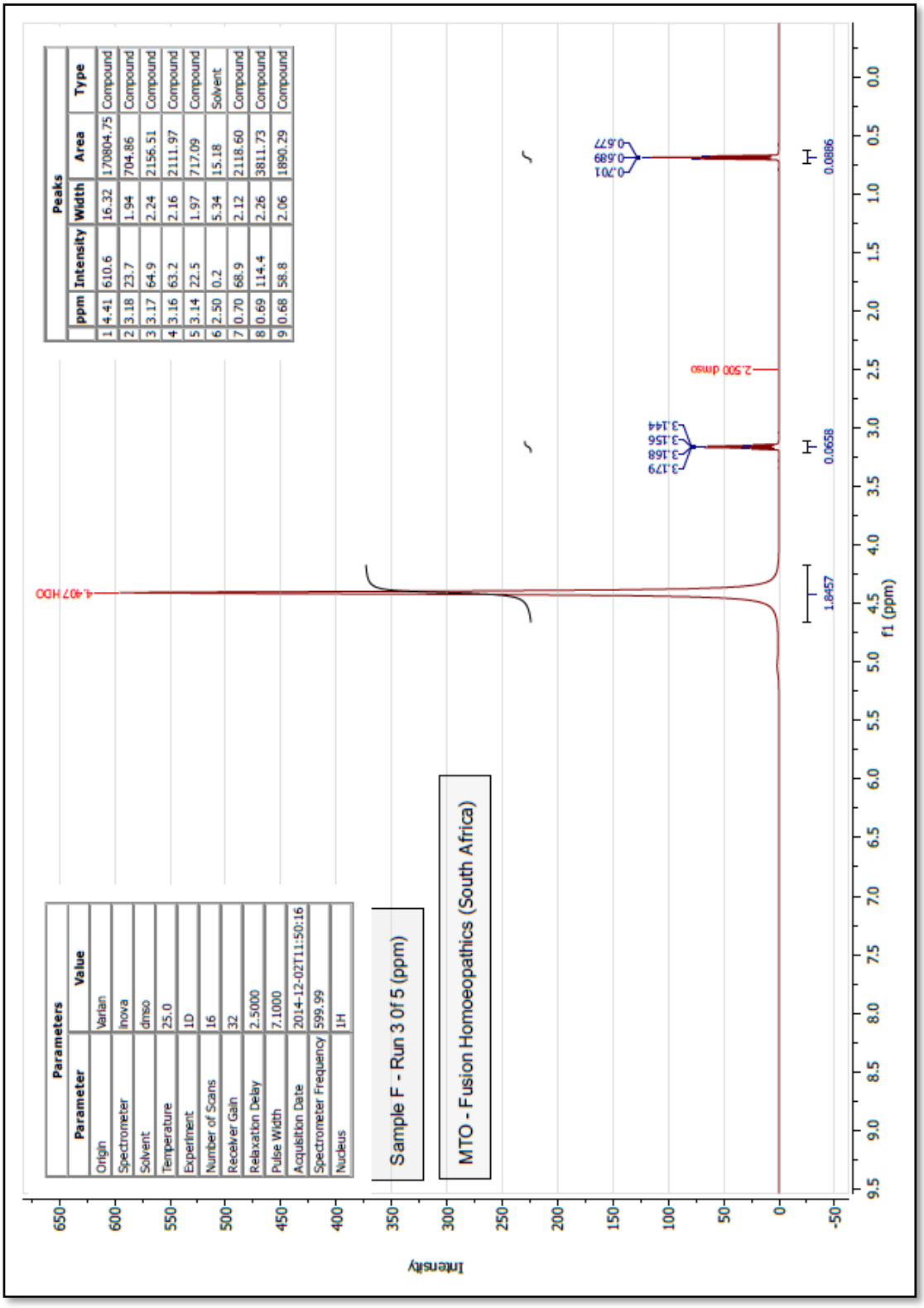
Graph 24: Sample E (Run 5 of 5) CONTROL - Lactose LM1



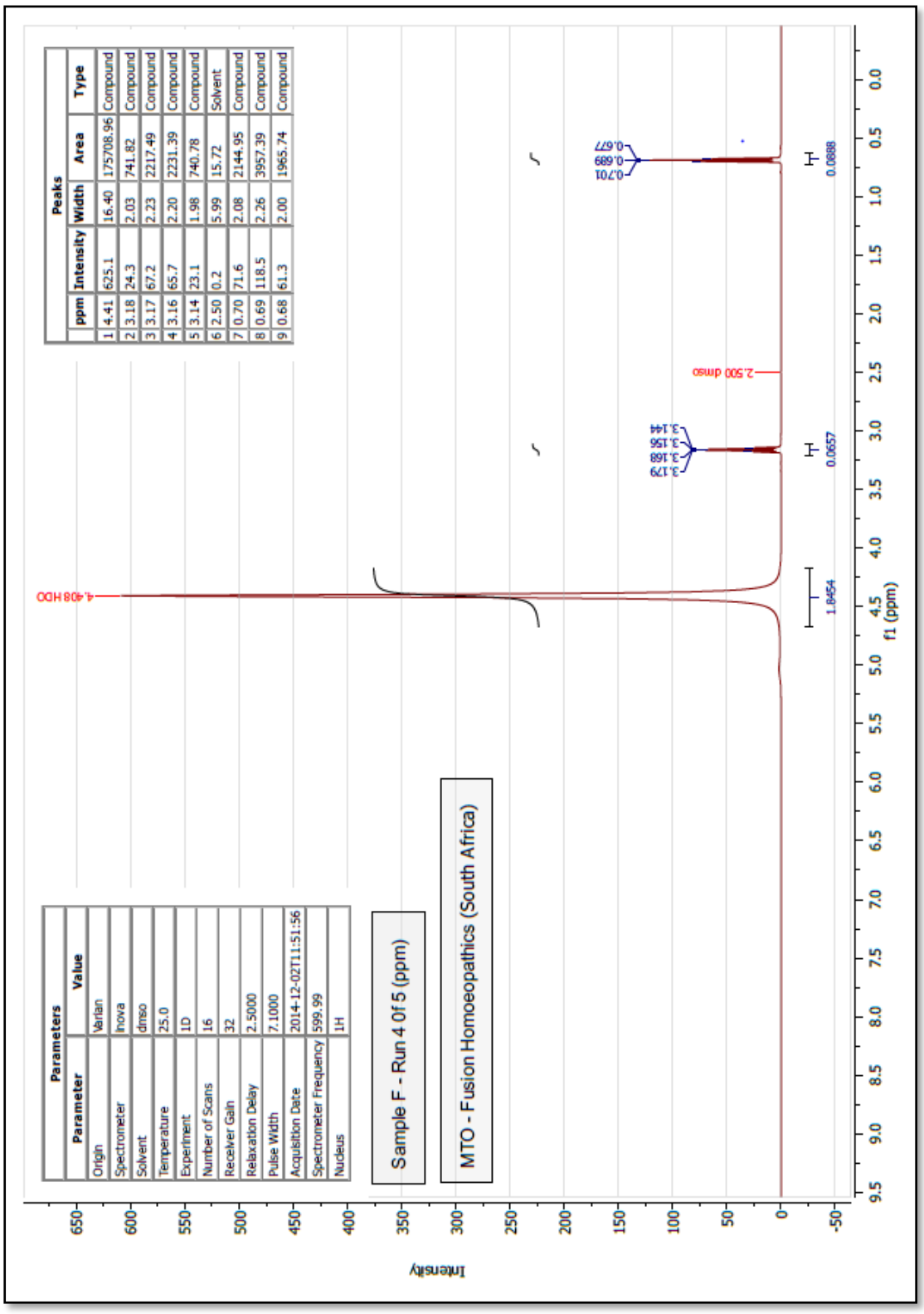
Graph 25: Sample F (Run 1 of 5) MTO - Fusion Homoeopathics (ZA)



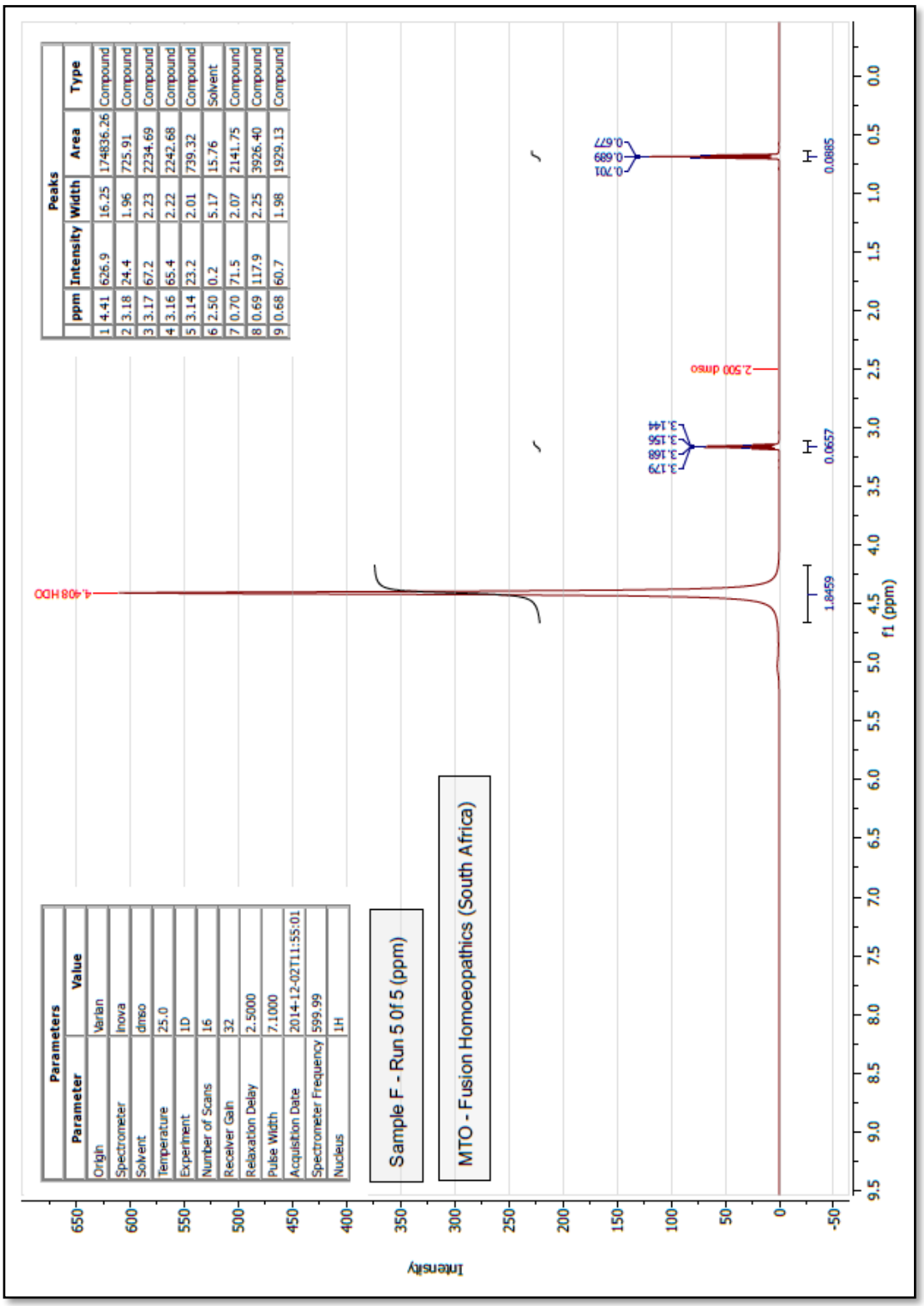
Graph 26: Sample F (Run 2 of 5) MTO - Fusion Homoeopathics (ZA)



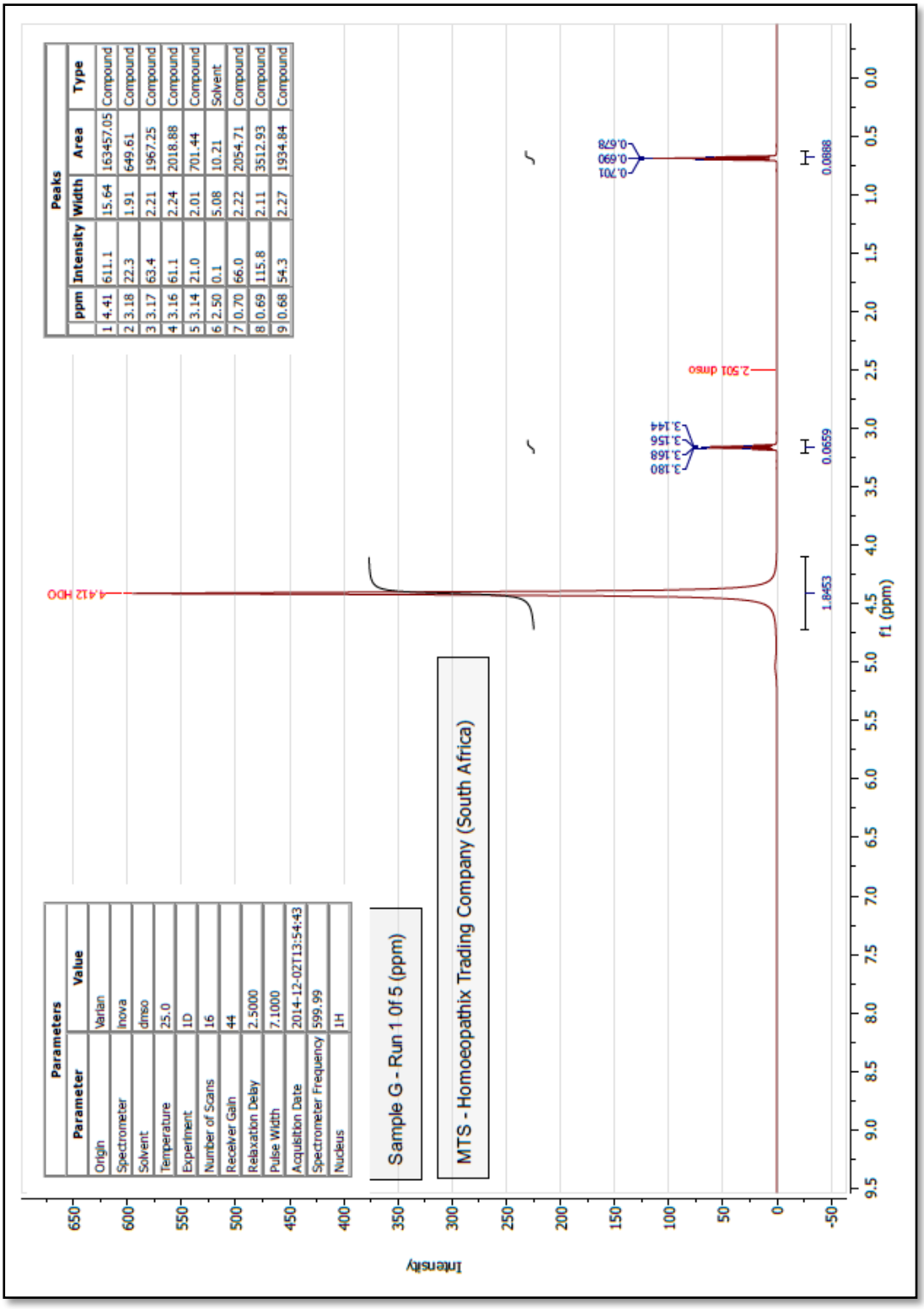
Graph 27: Sample F (Run 3 of 5) MTO - Fusion Homoeopathics (ZA)



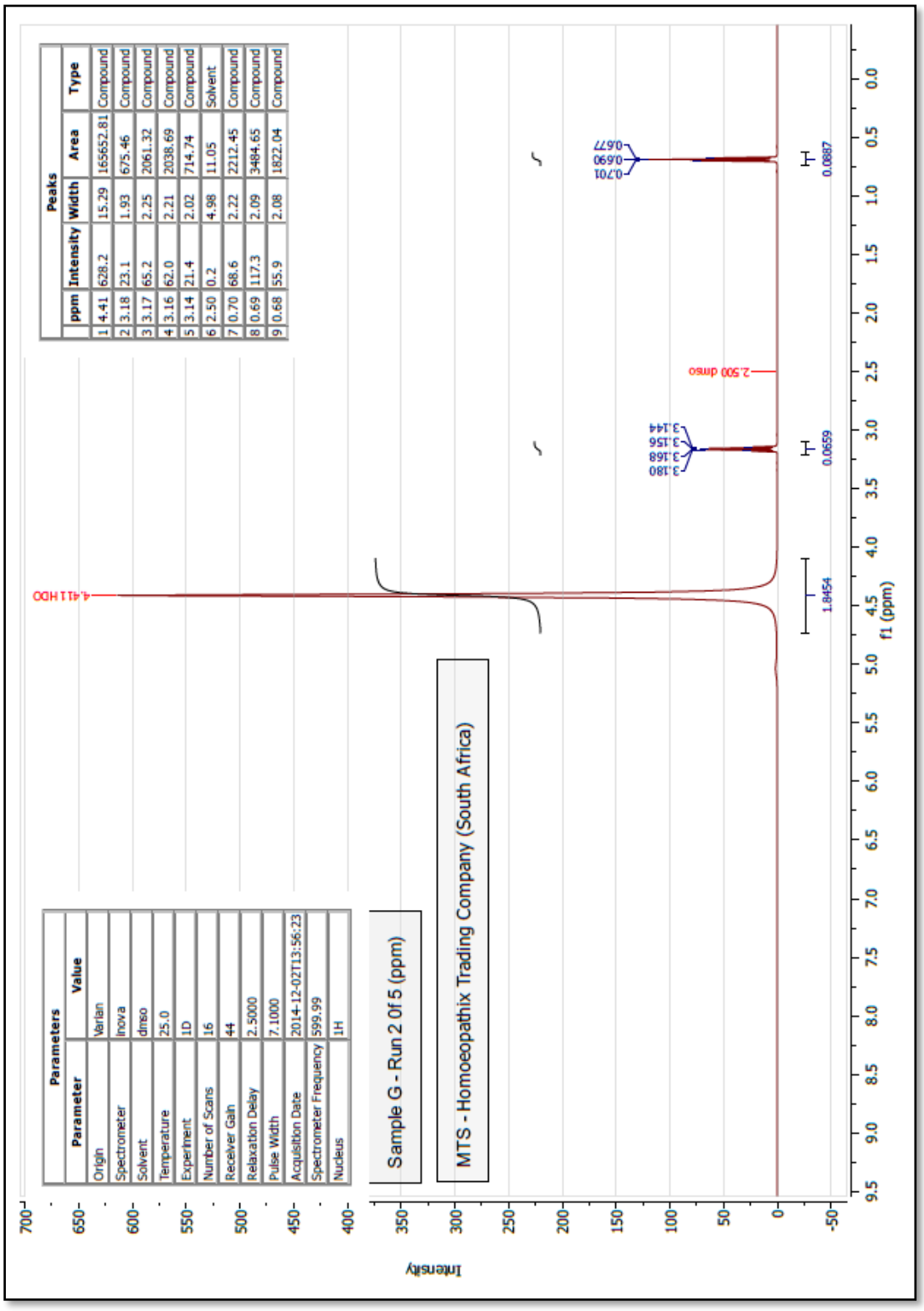
Graph 28: Sample F (Run 4 of 5) MTO - Fusion Homoeopathics (ZA)



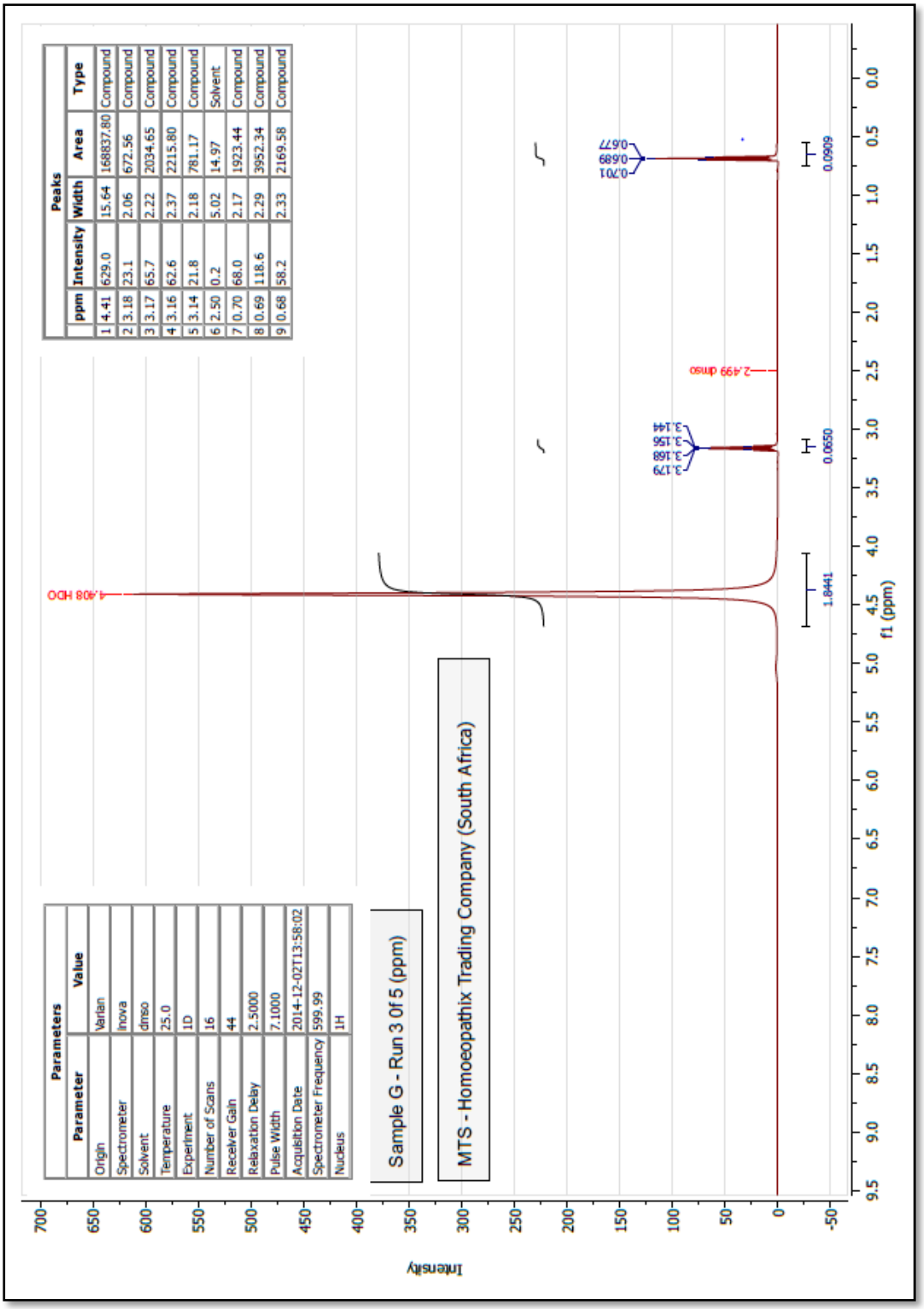
Graph 29: Sample F (Run 5 of 5) MTO - Fusion Homoeopathics (ZA)



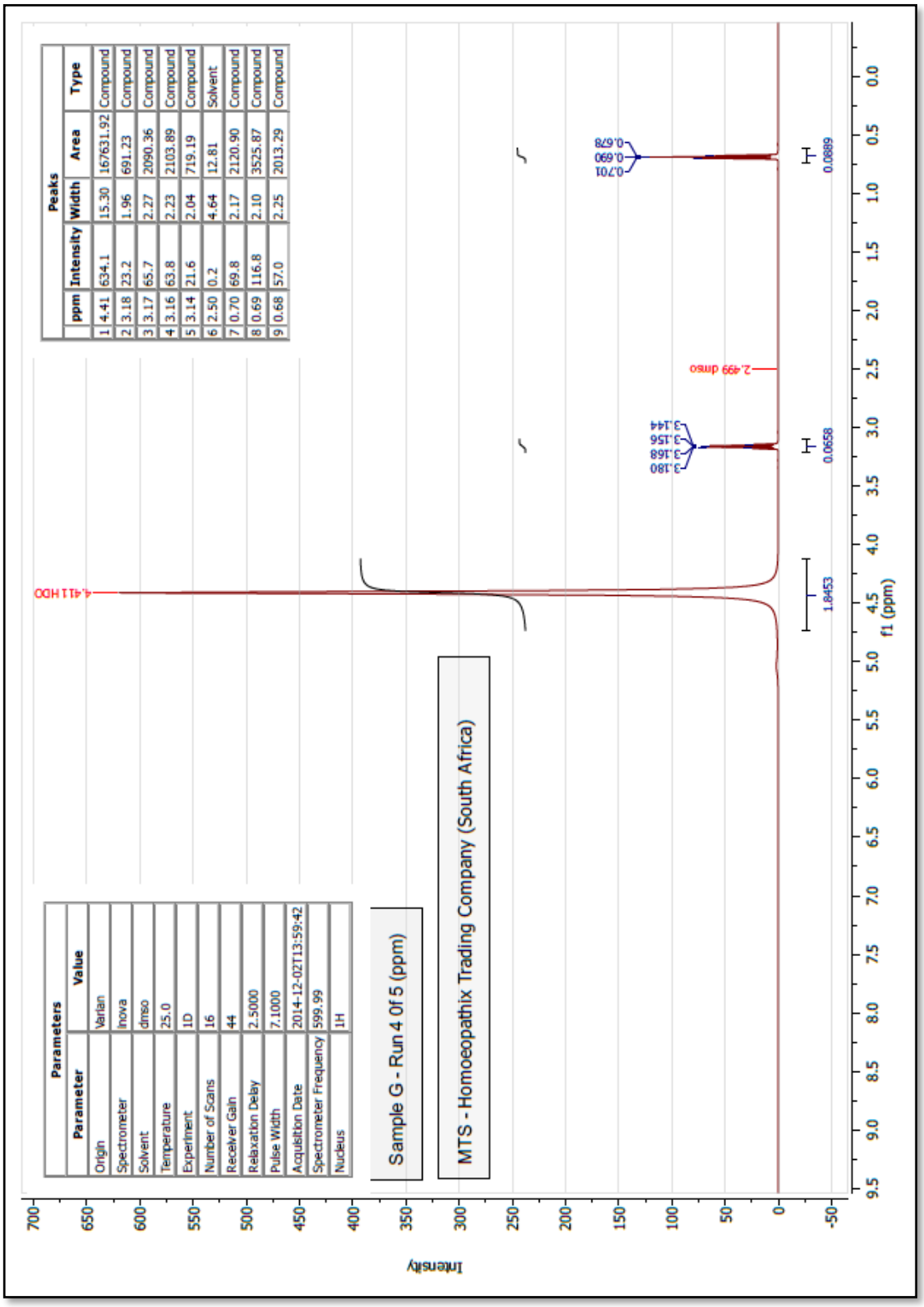
Graph 30: Sample G (Run 1 of 5) MTS - Homoeopathix Trading Co. (ZA)



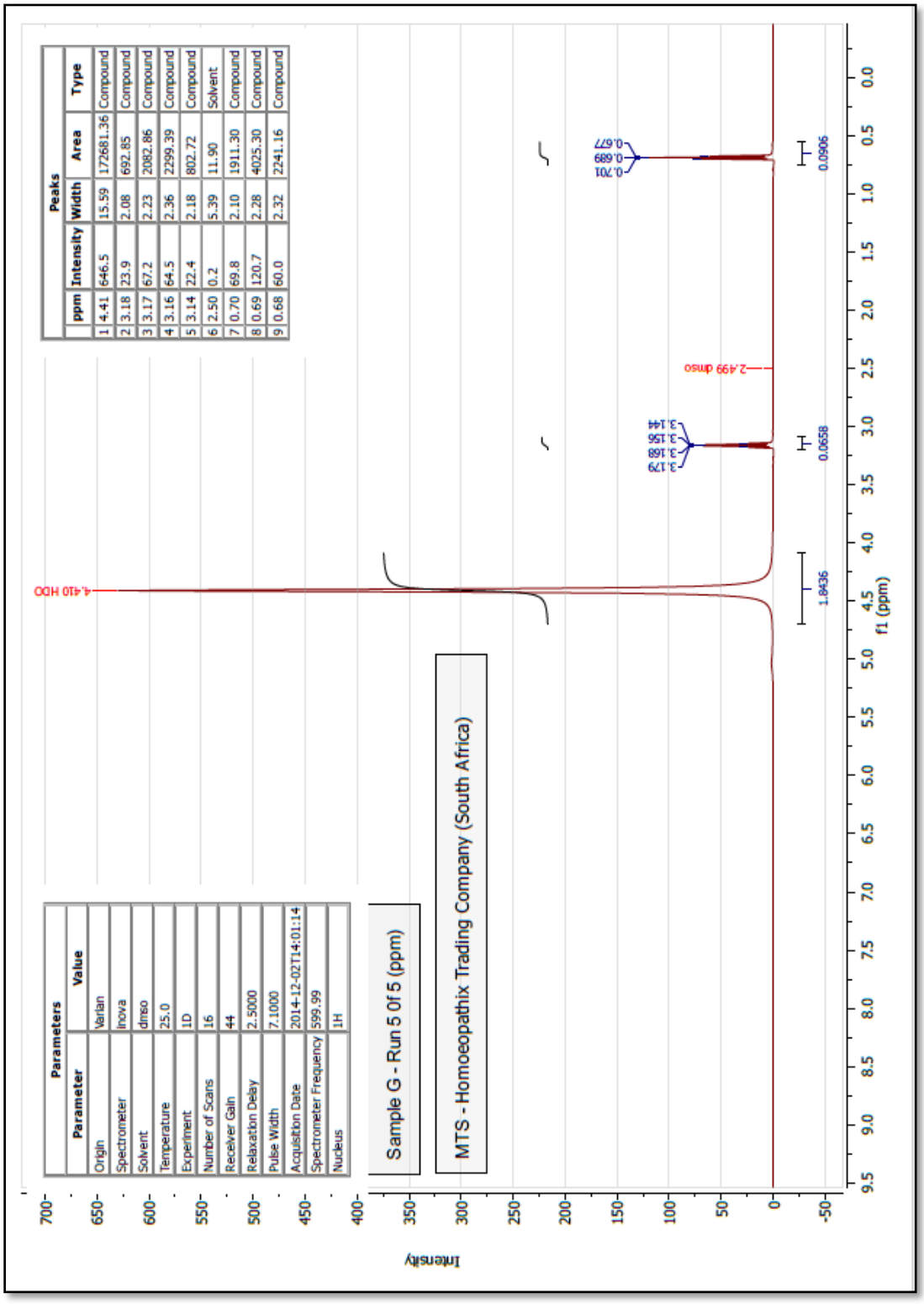
Graph 31: Sample G (Run 2 of 5) MTS - Homoeopathix Trading Co. (ZA)



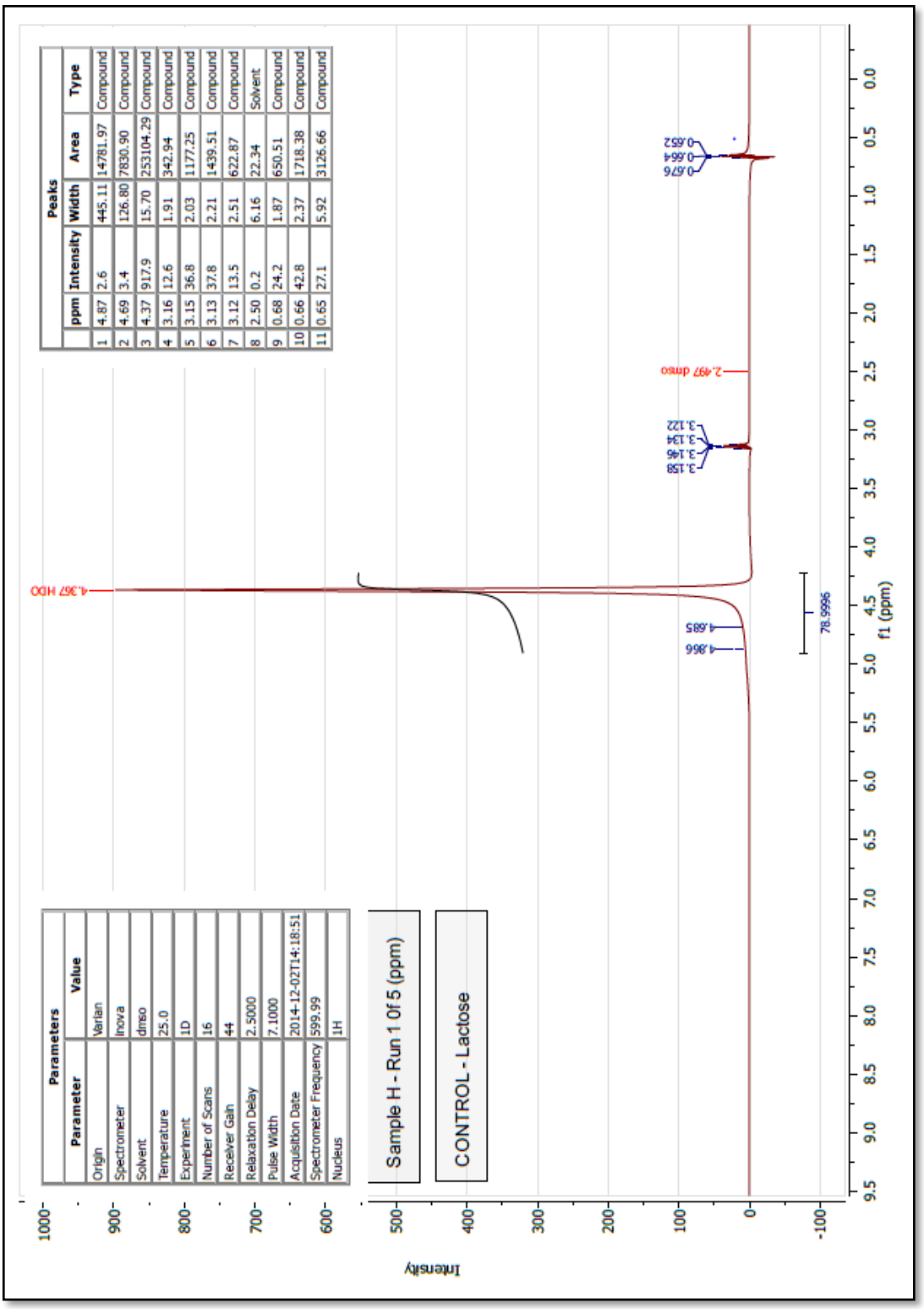
Graph 32: Sample G (Run 3 of 5) MTS - Homoeopathix Trading Co. (ZA)



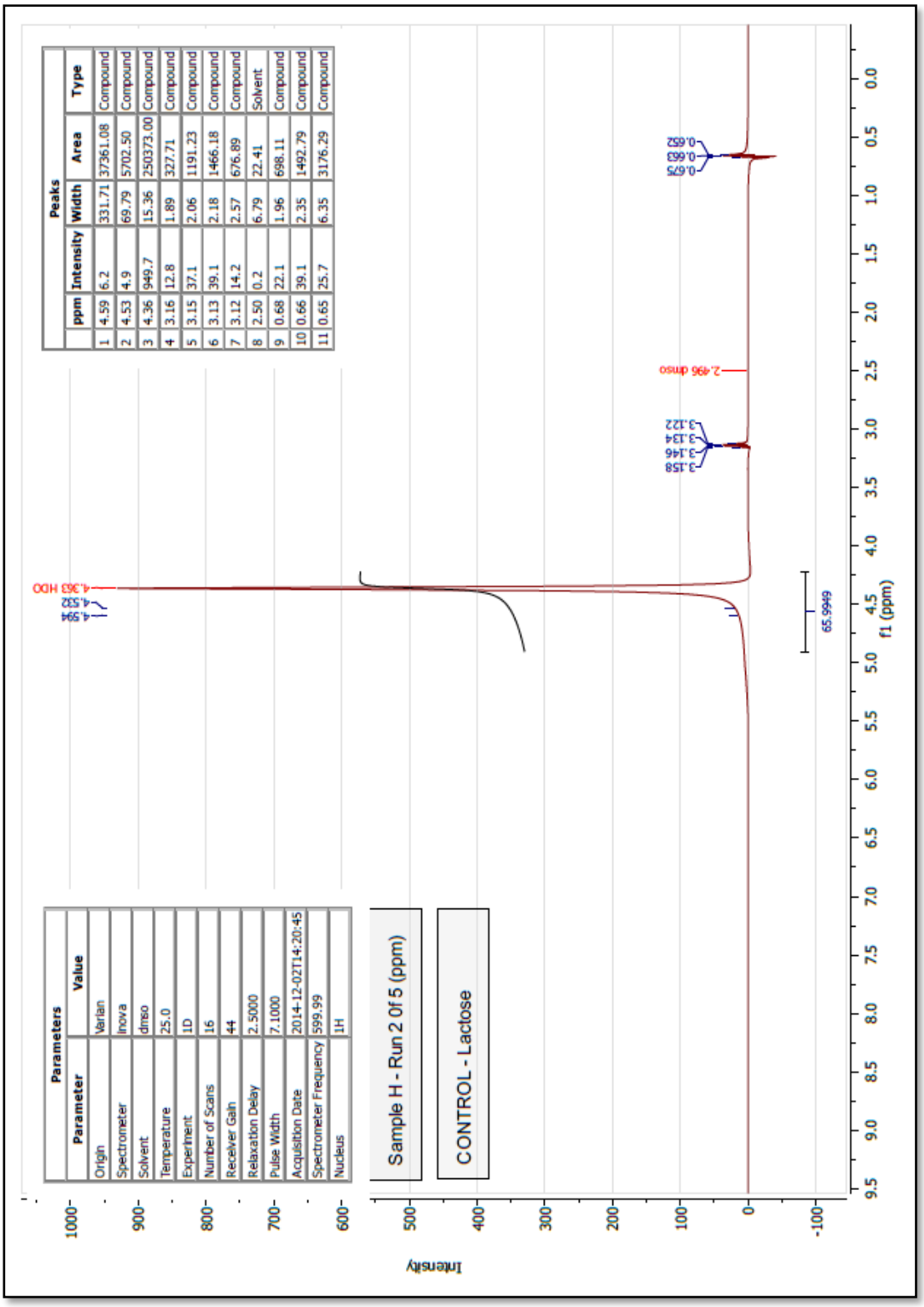
Graph 33: Sample G (Run 4 of 5) MTS - Homoeopathix Trading Co. (ZA)



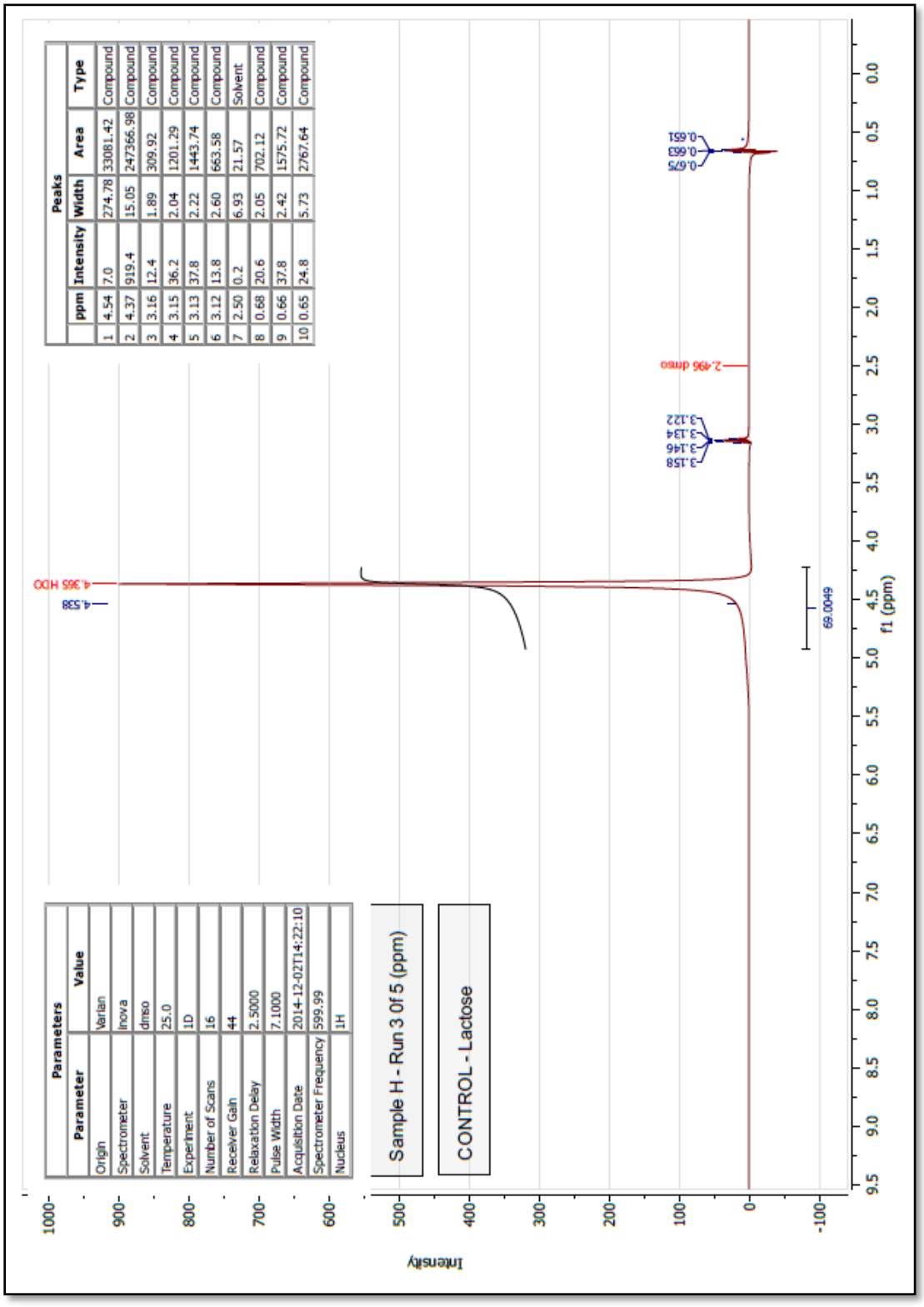
Graph 34: Sample G (Run 5 of 5) MTS - Homoeopathix Trading Co. (ZA)



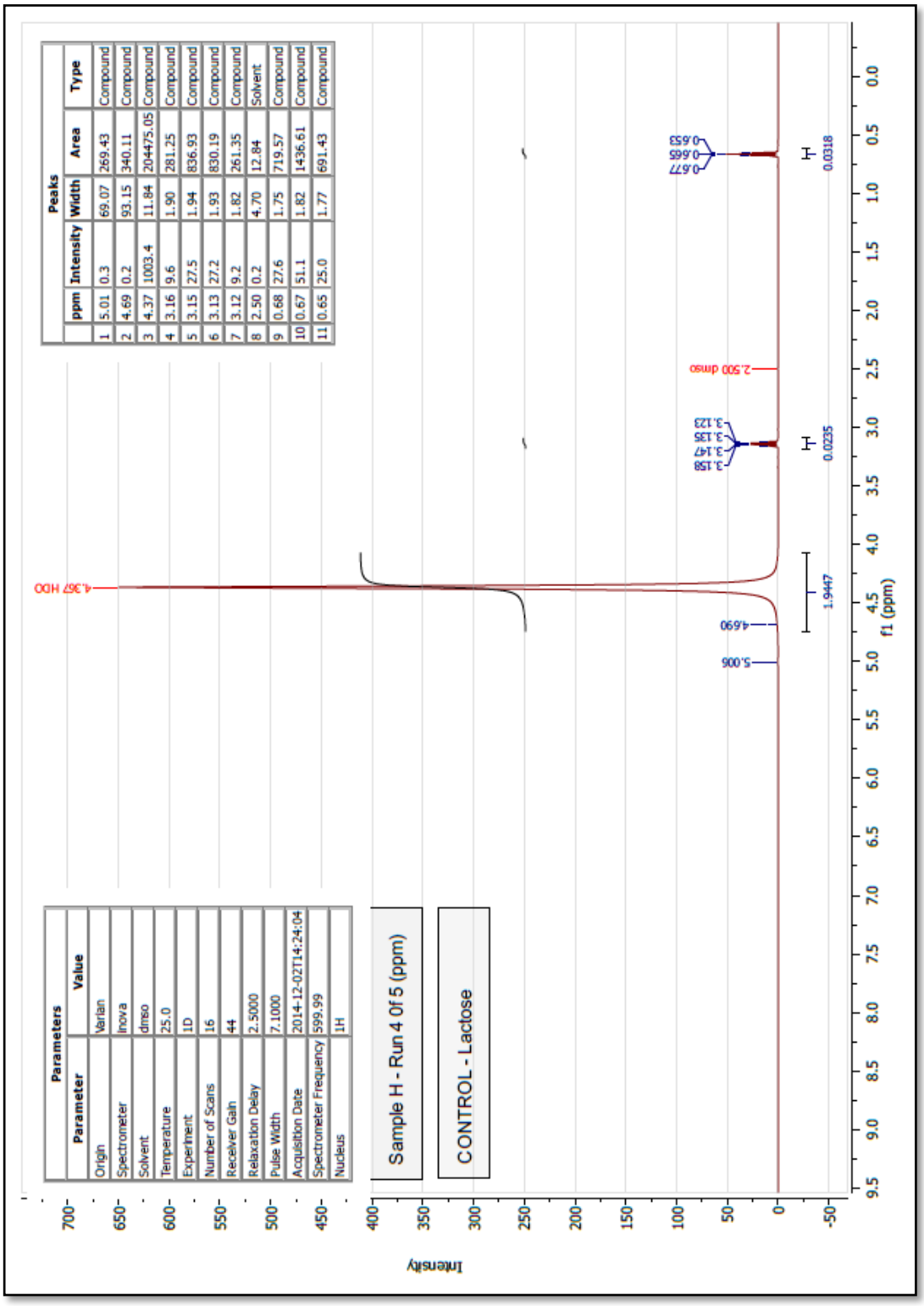
Graph 35: Sample H (Run 1 of 5) CONTROL - Lactose



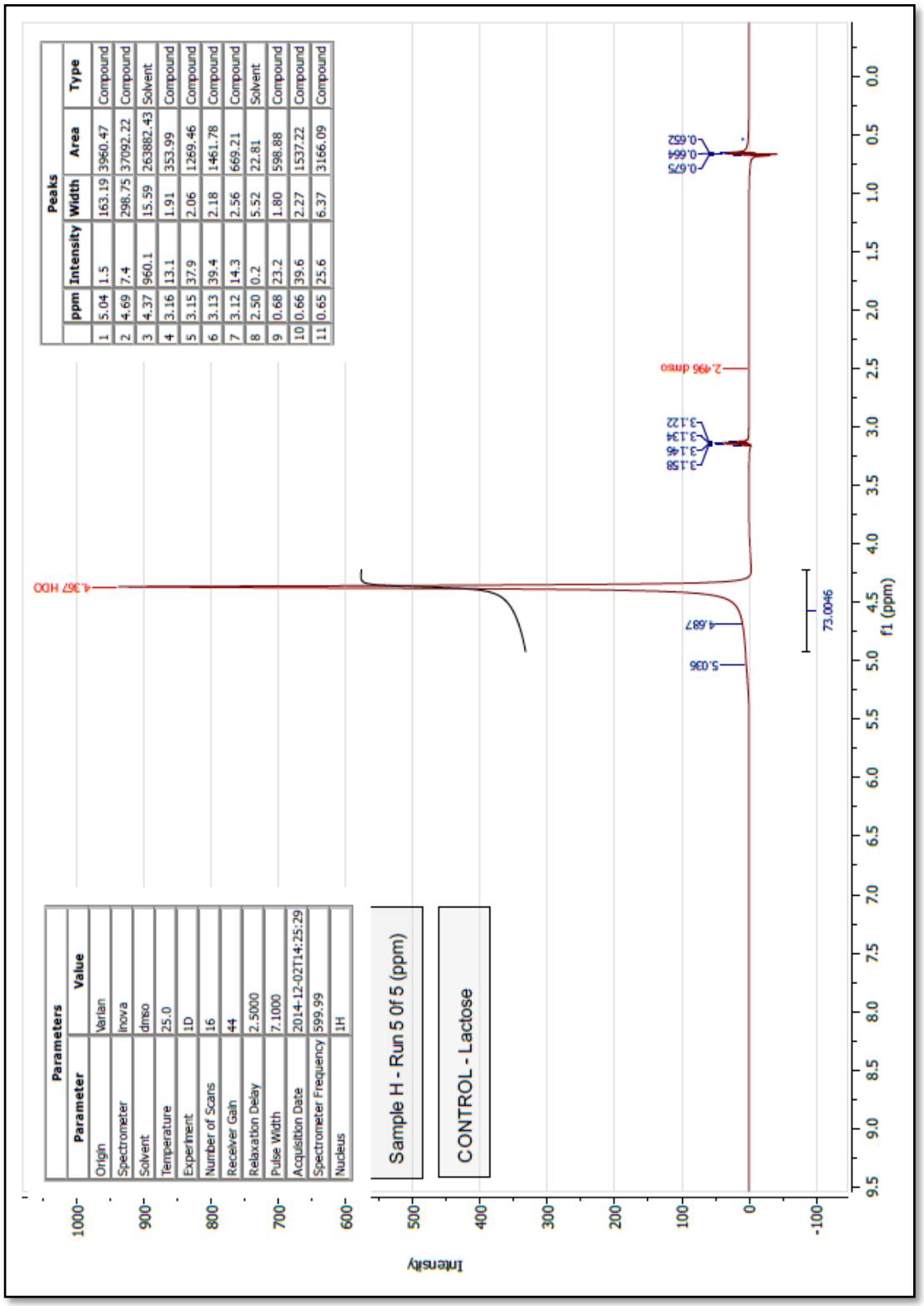
Graph 36: Sample H (Run 2 of 5) CONTROL - Lactose



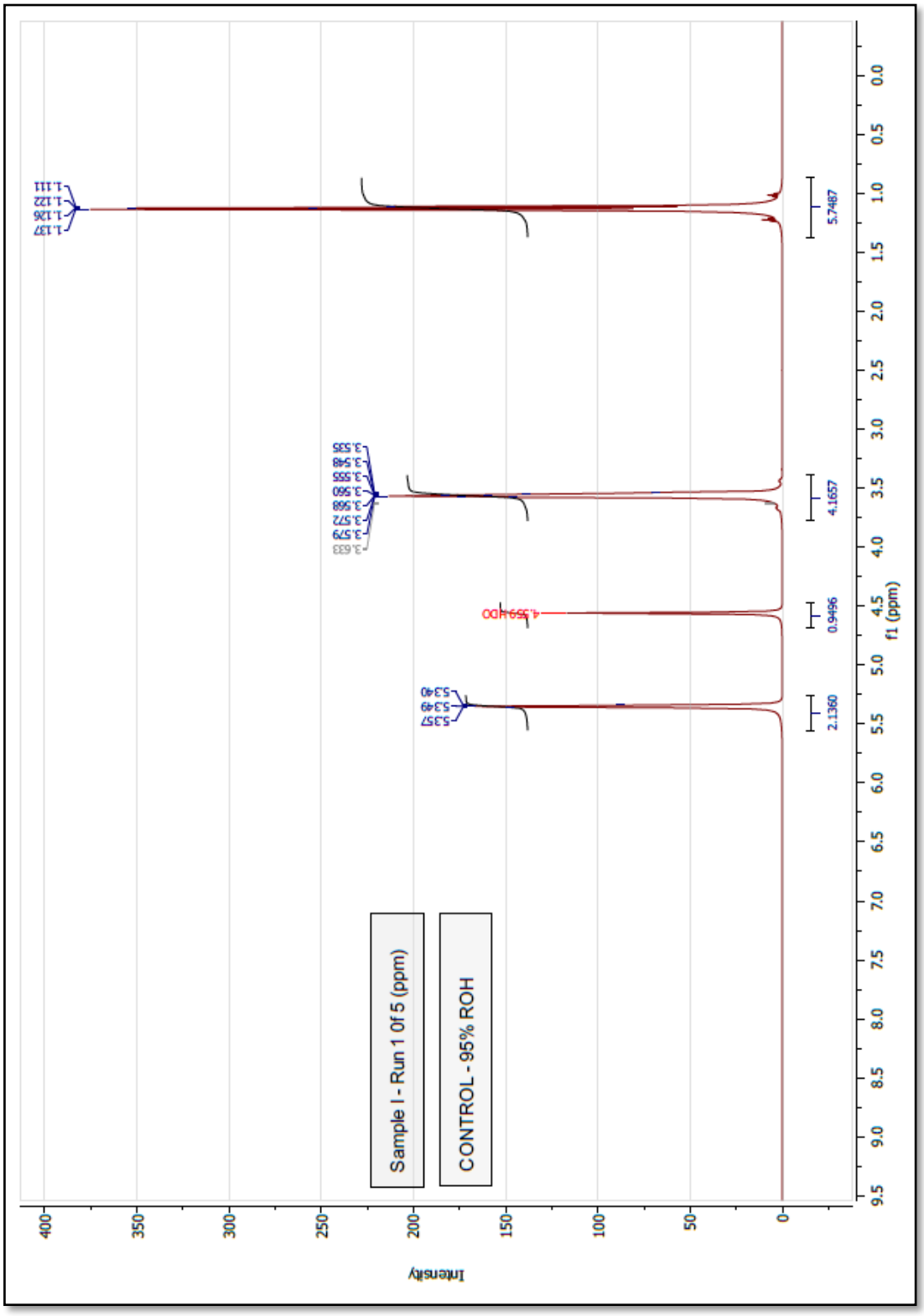
Graph 37: Sample H (Run 3 of 5) CONTROL - Lactose



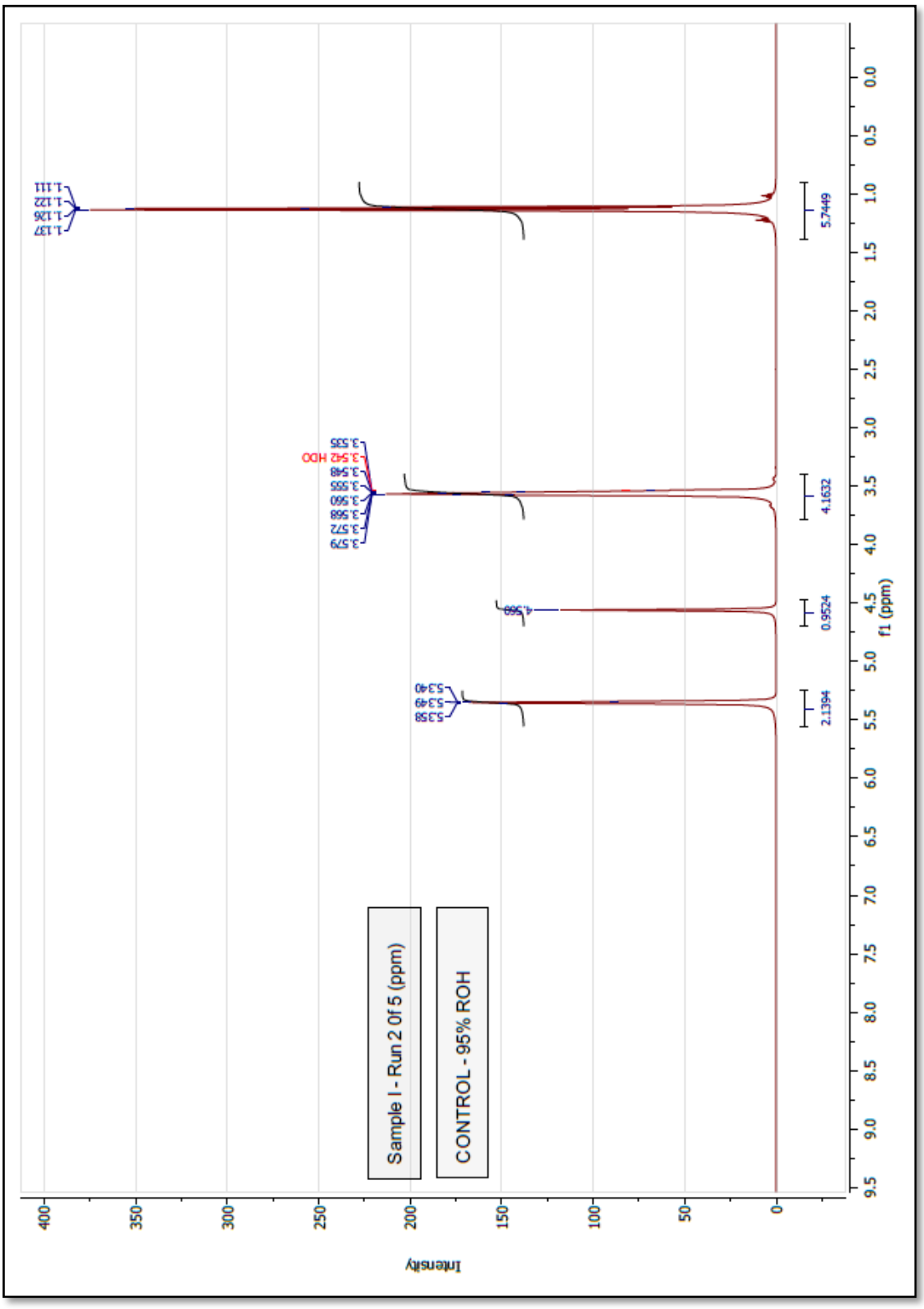
Graph 38: Sample H (Run 4 of 5) CONTROL - Lactose



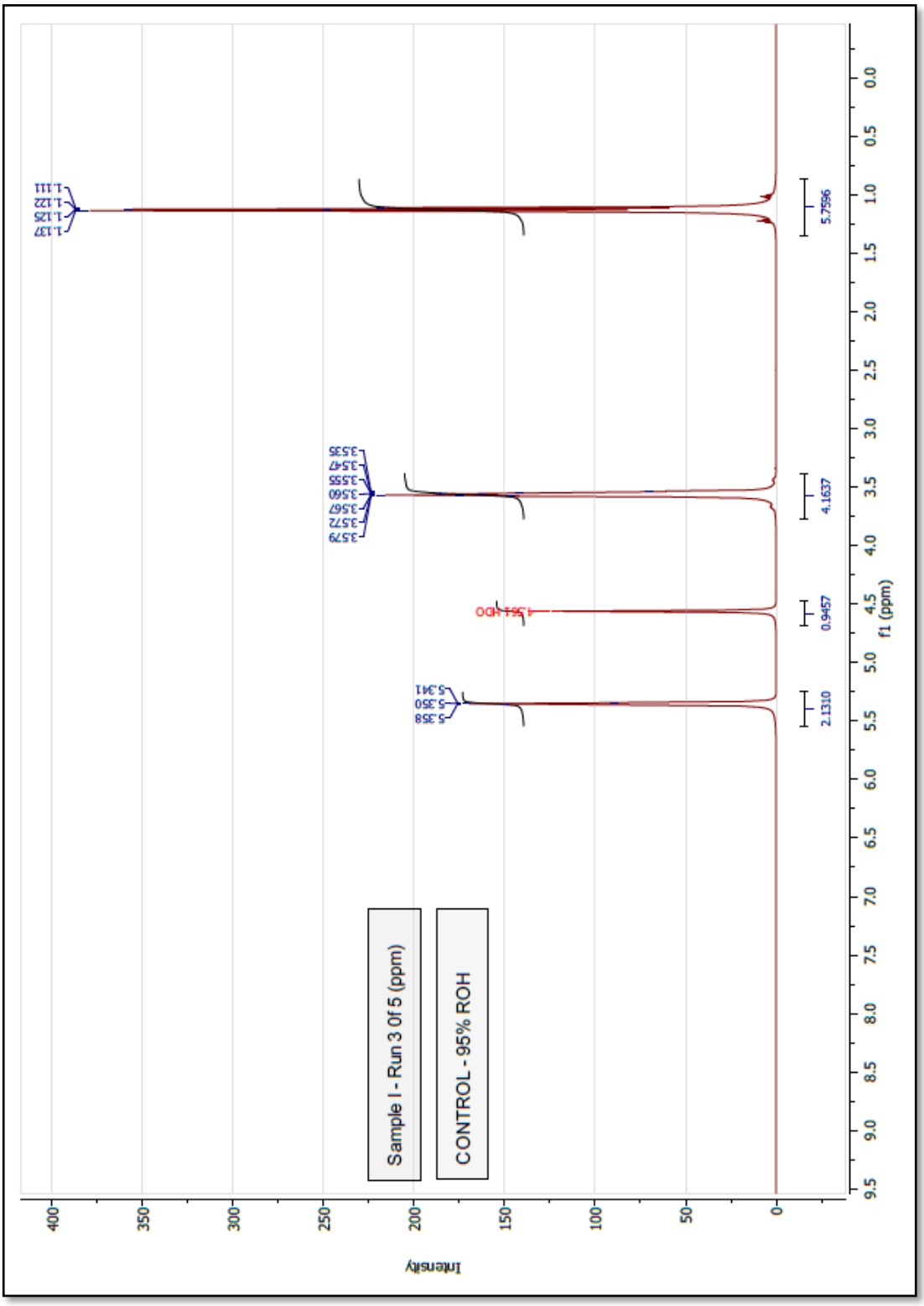
Graph 39: Sample H (Run 5 of 5) CONTROL - Lactose



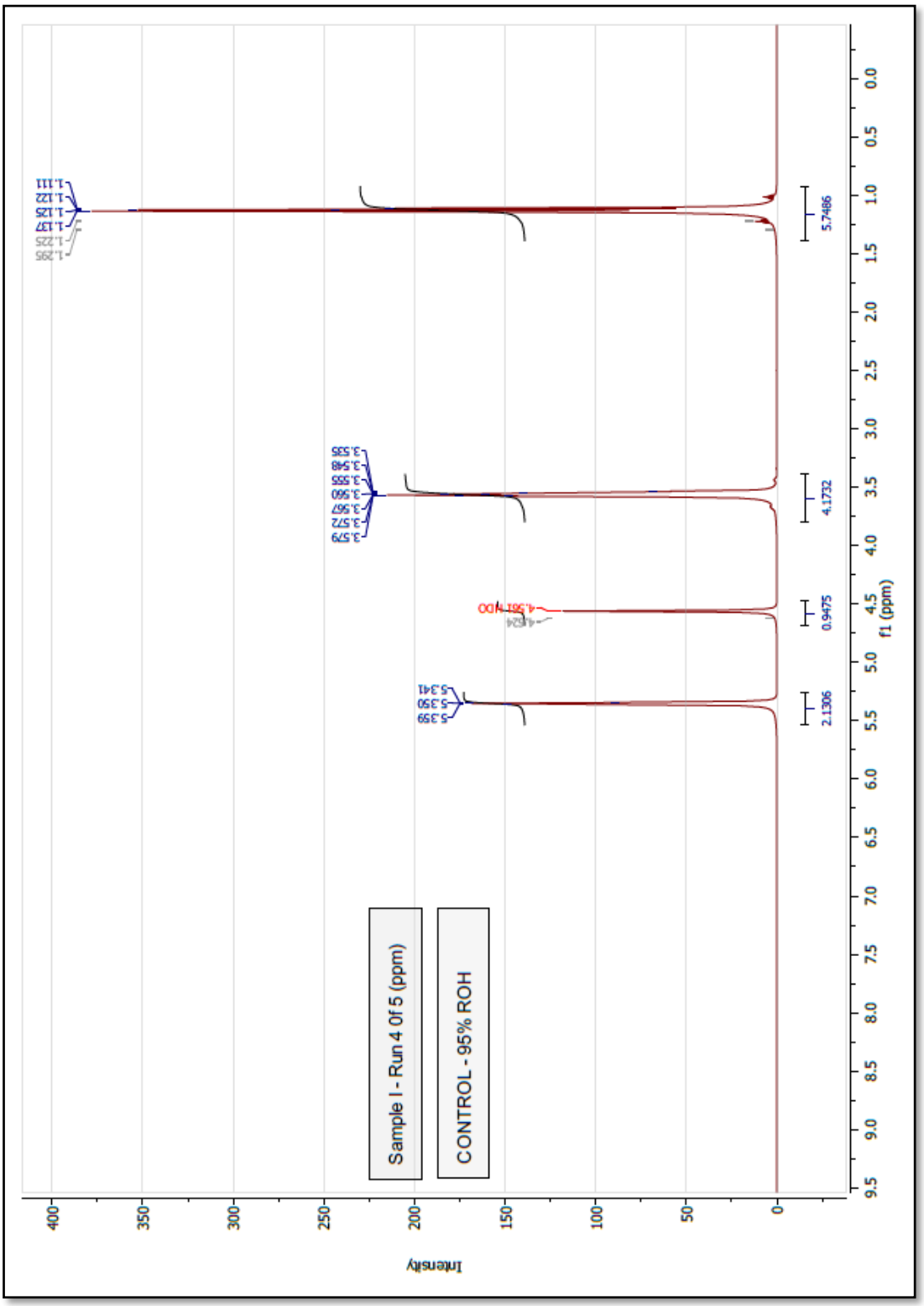
Graph 40: Sample I (Run 1 of 5) CONTROL - 95% ROH



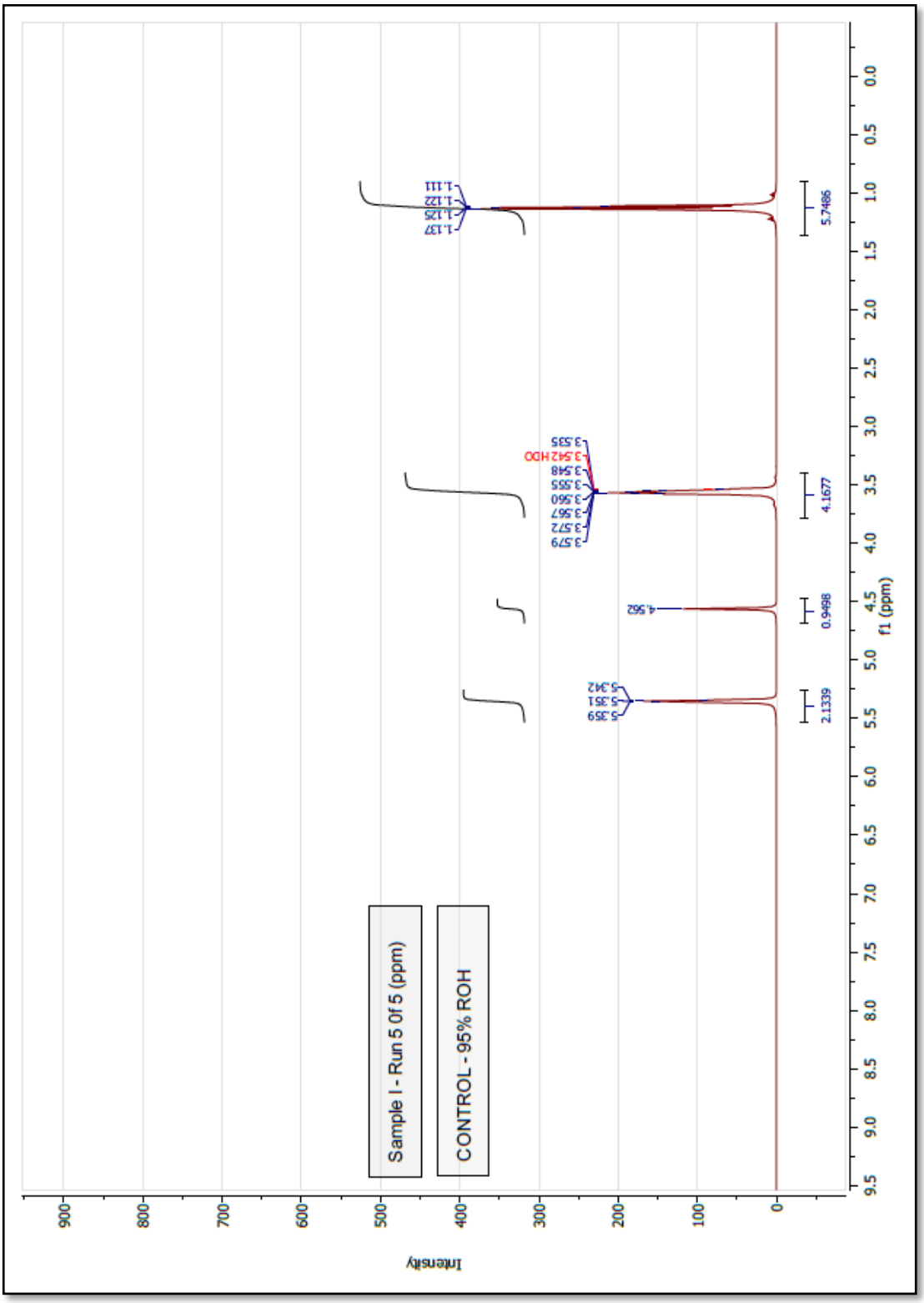
Graph 41: Sample I (Run 2 of 5) CONTROL - 95% ROH



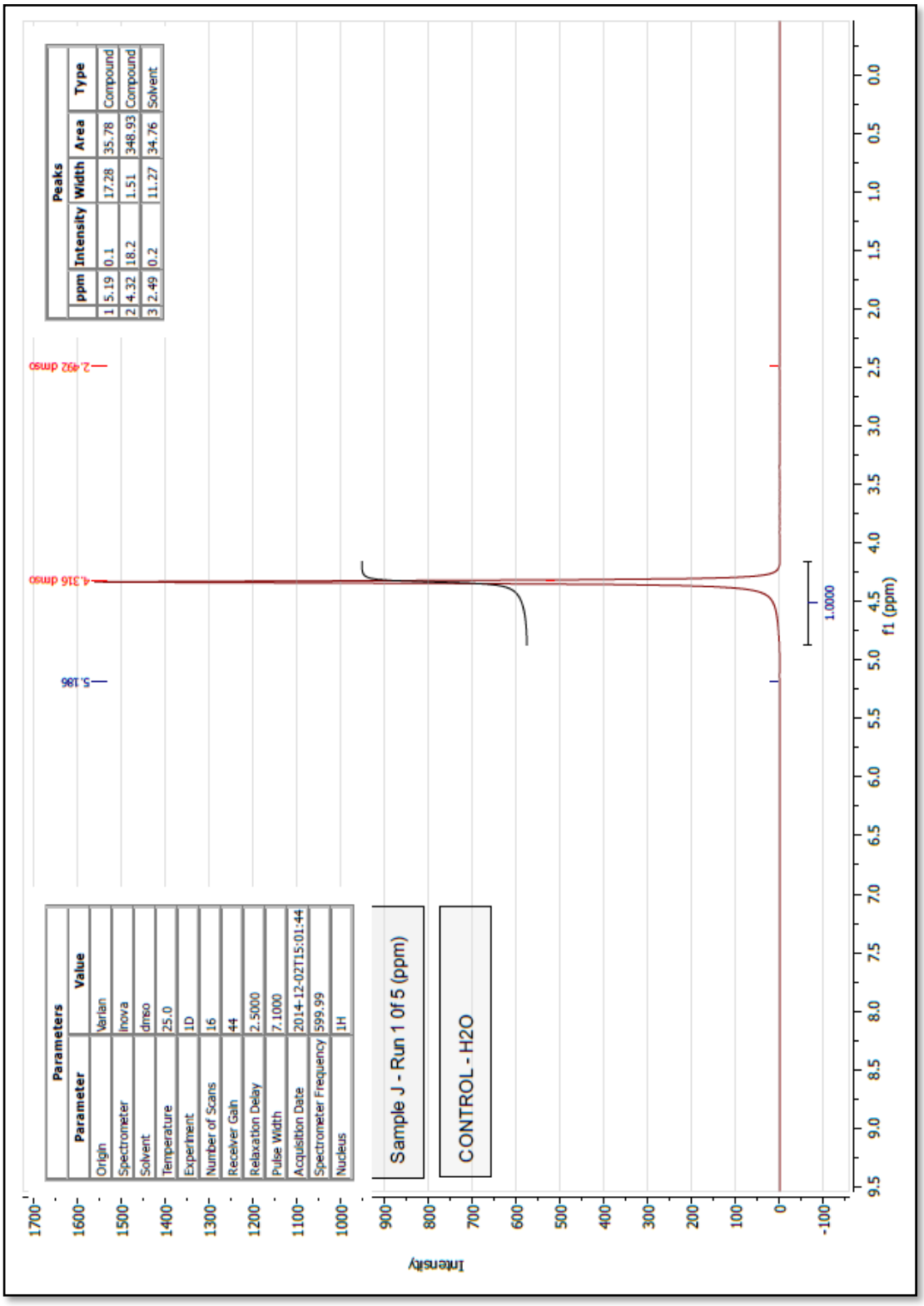
Graph 42: Sample I (Run 3 of 5) CONTROL - 95% ROH



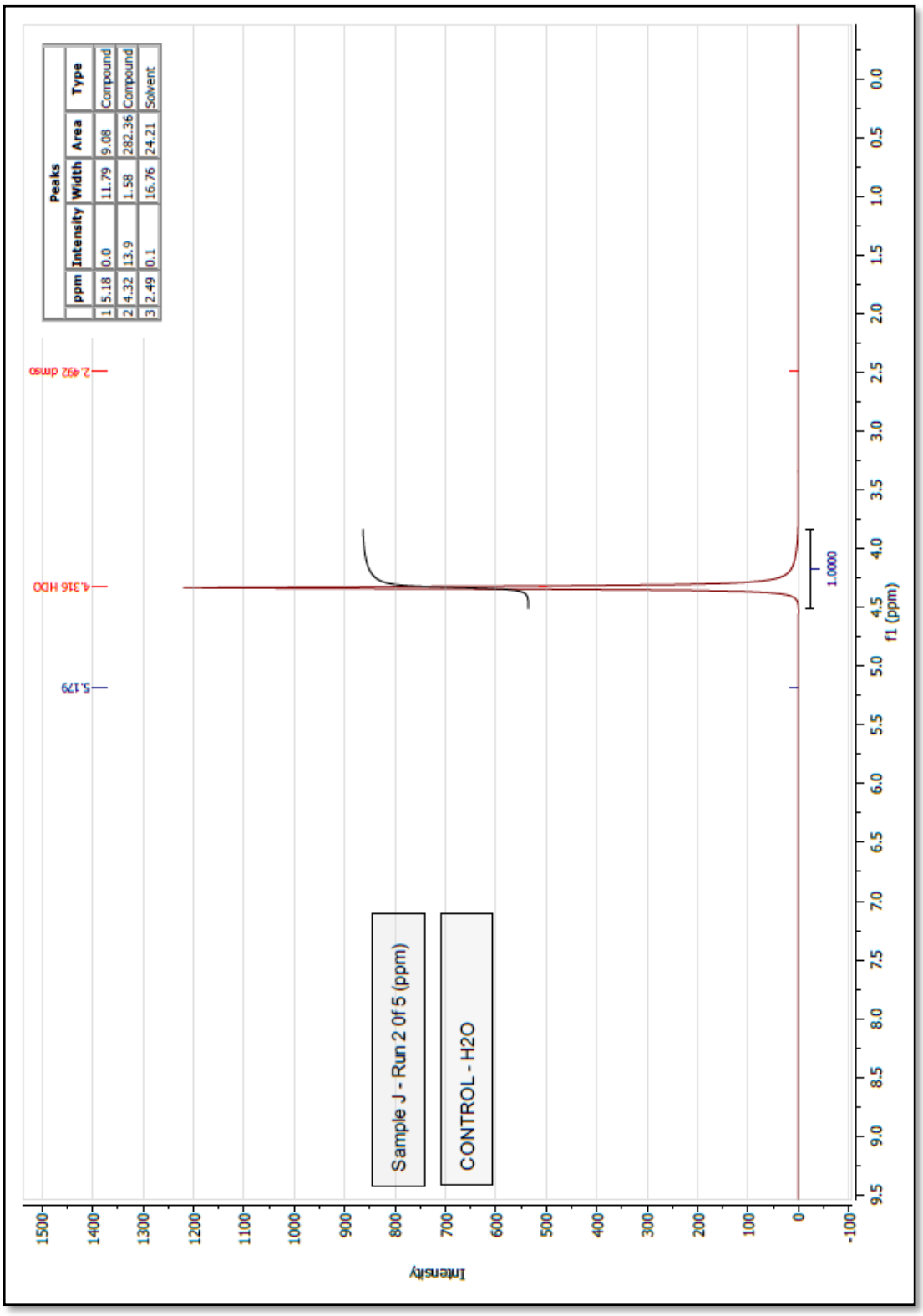
Graph 43: Sample I (Run 4 of 5) CONTROL - 95% ROH



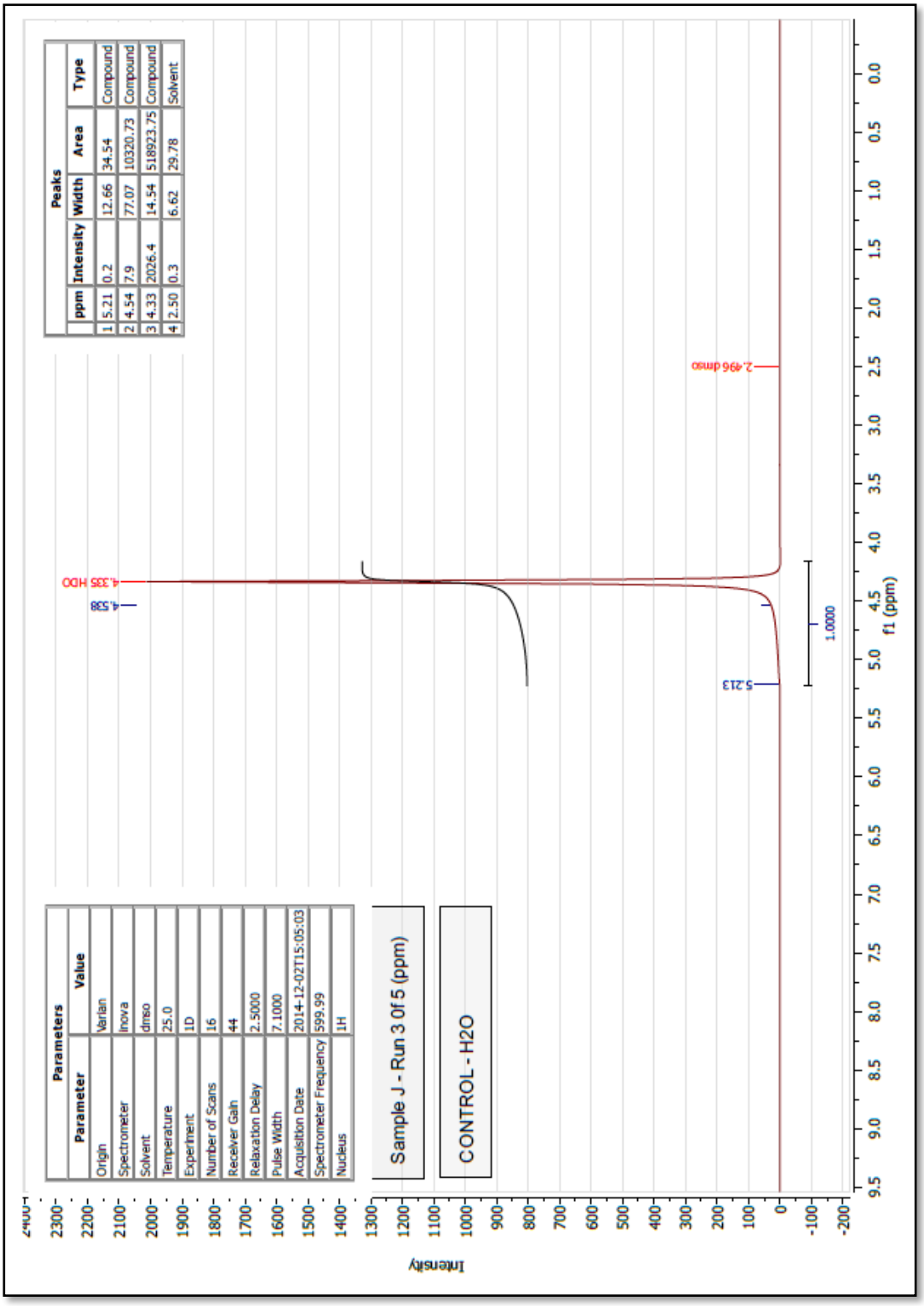
Graph 44: Sample I (Run 5 of 5) CONTROL - 95% ROH



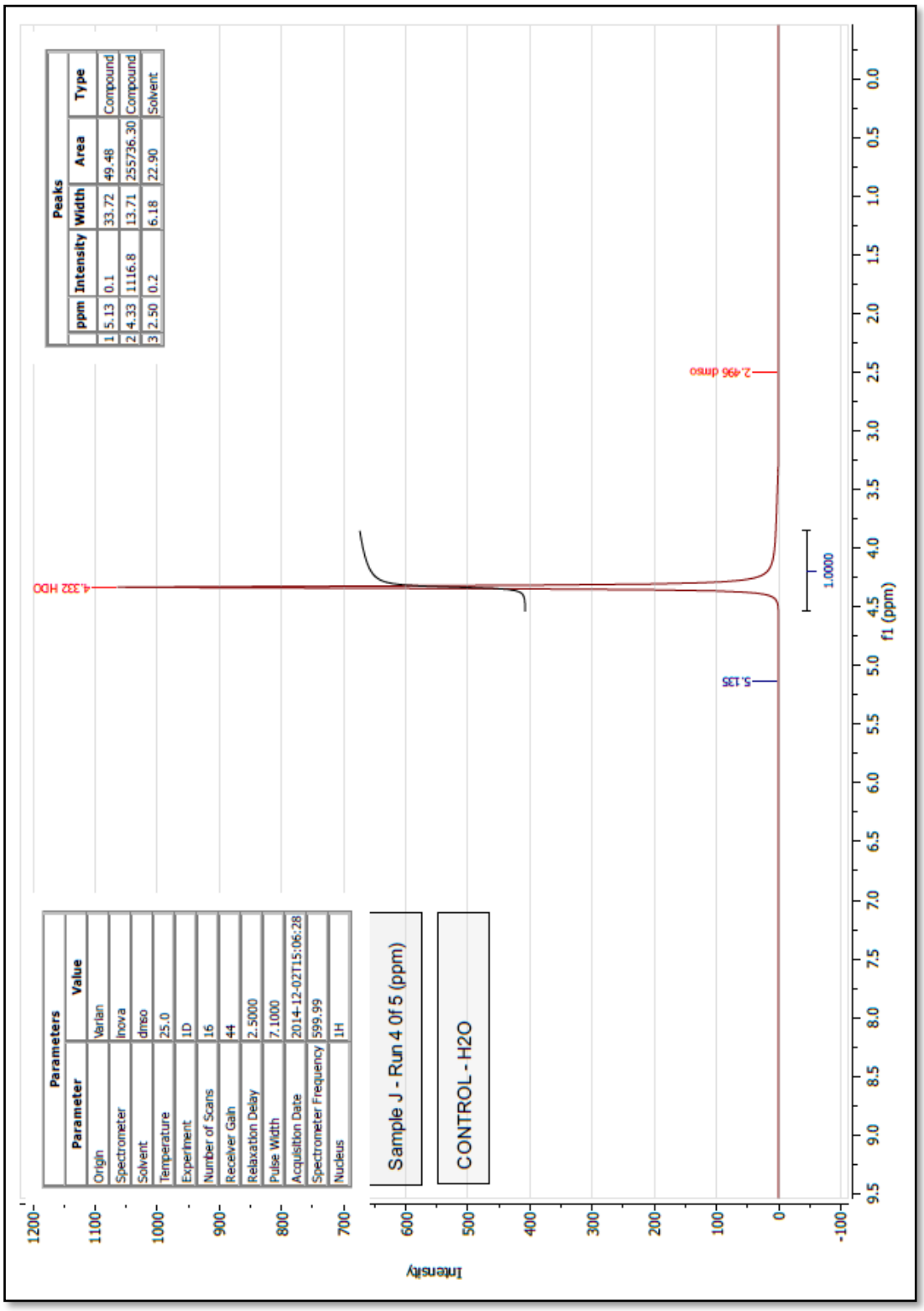
Graph 45: Sample J (Run 1 of 5) CONTROL - H_2O



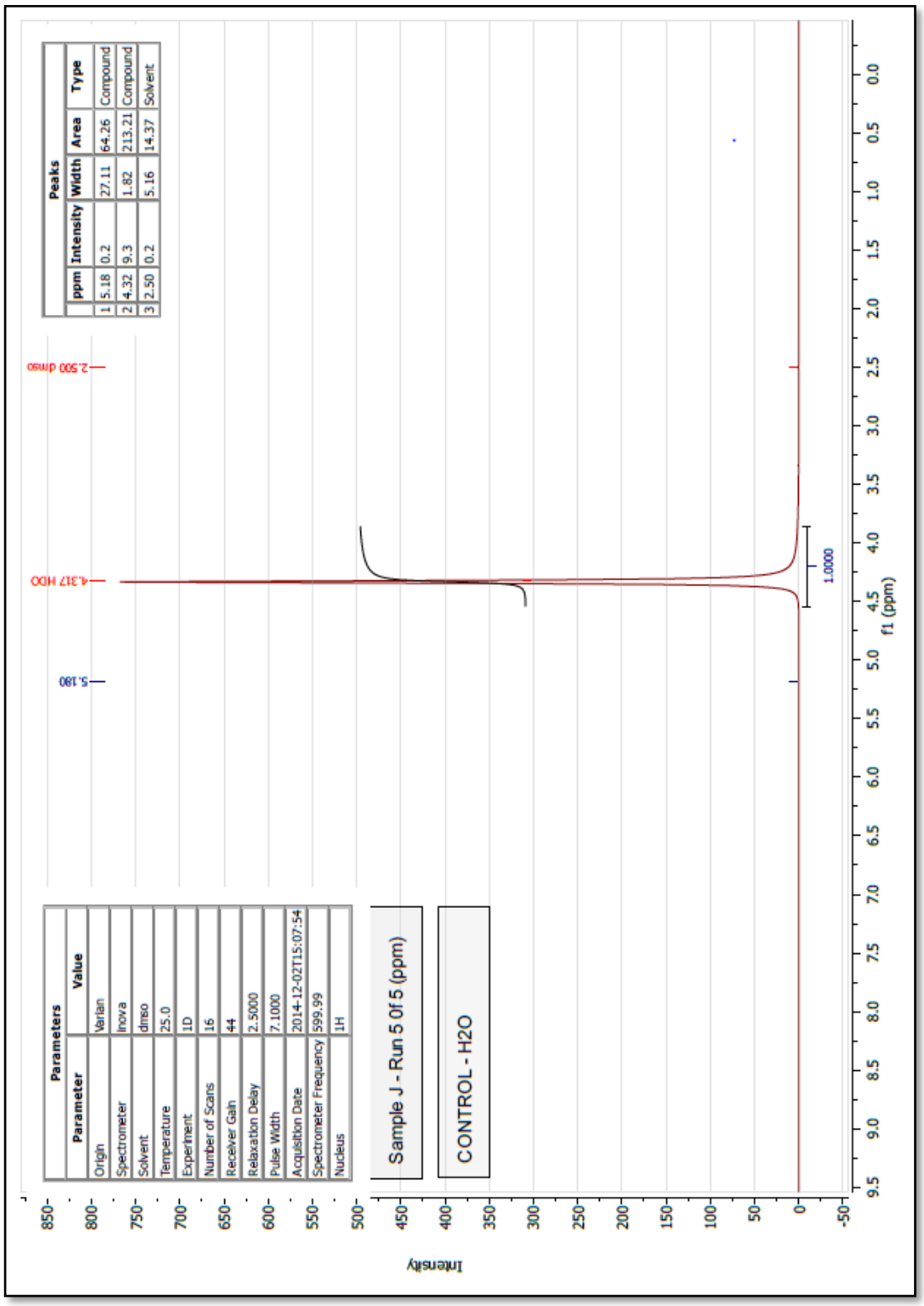
Graph 46: Sample J (Run 2 of 5) CONTROL - H_2O



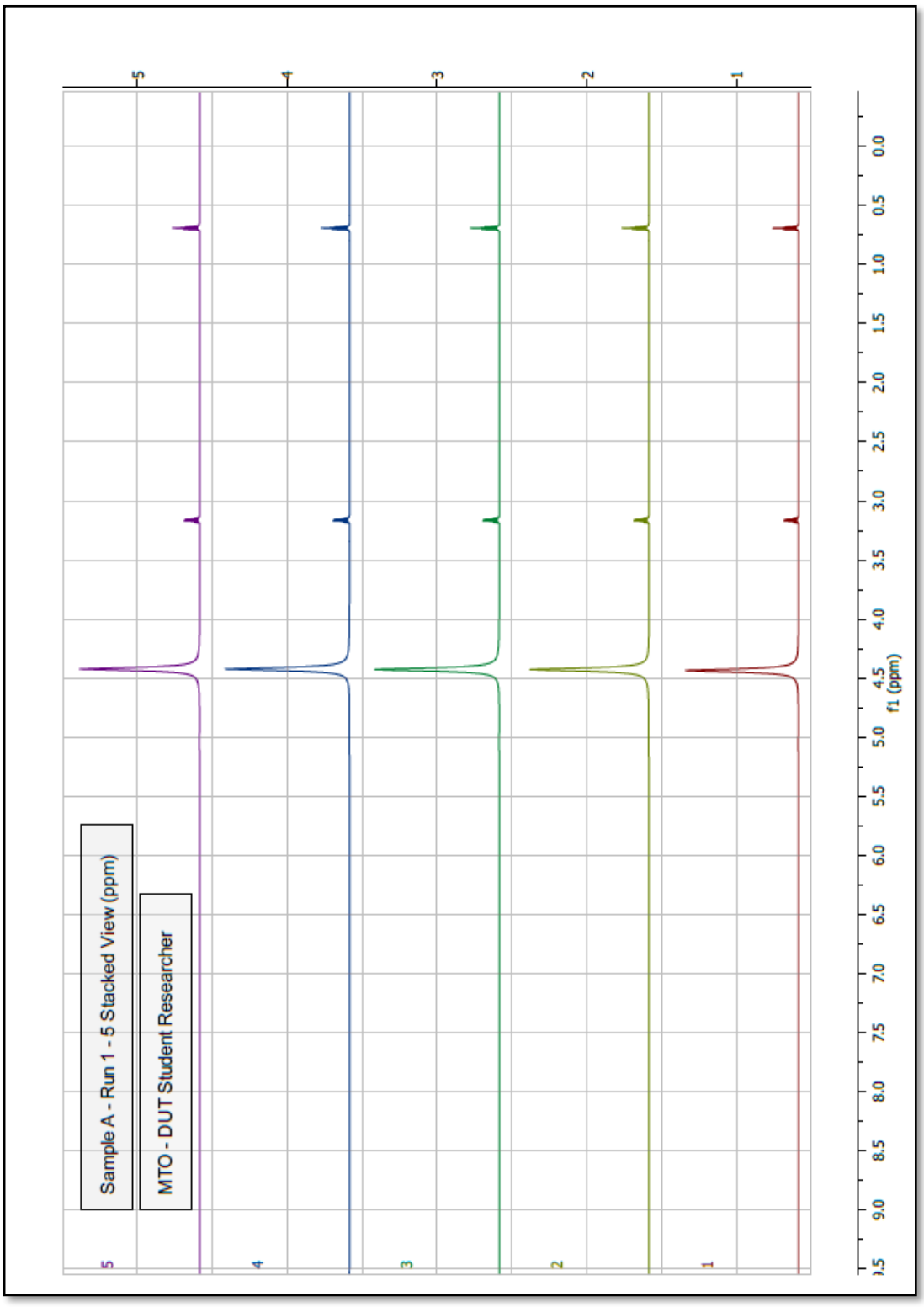
Graph 47: Sample J (Run 3 of 5) CONTROL - H_2O



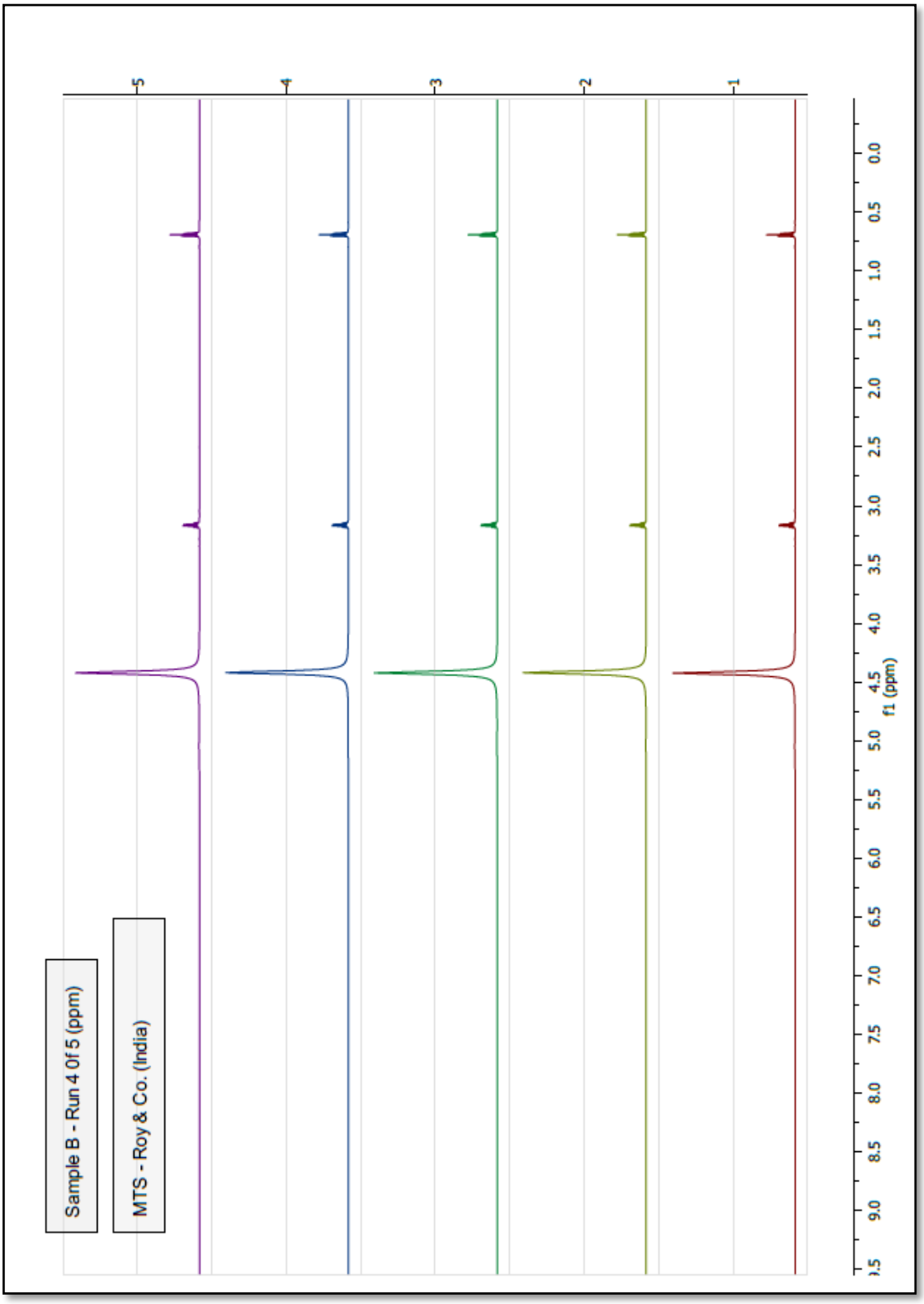
Graph 48: Sample J (Run 4 of 5) CONTROL - H_2O



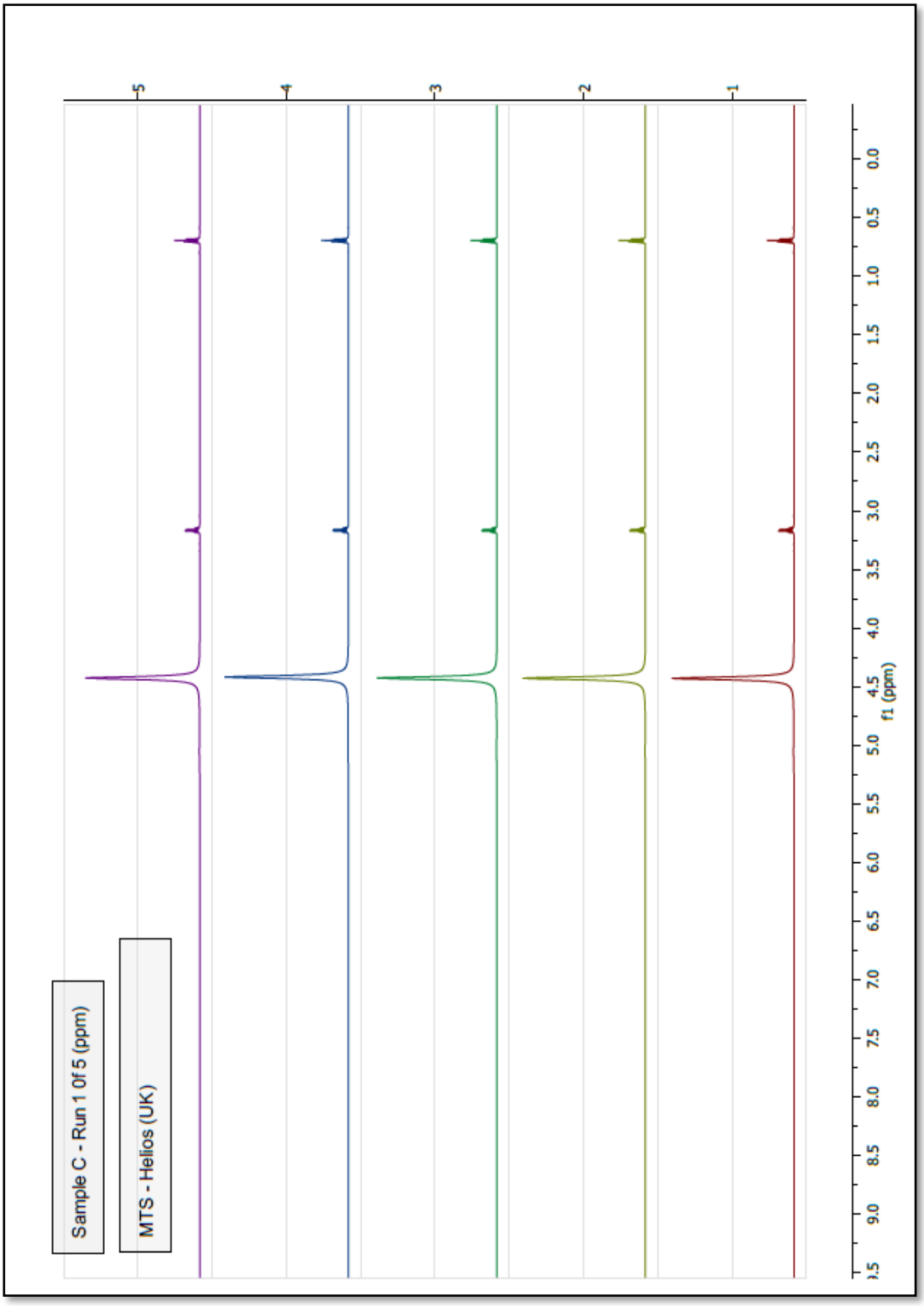
Graph 49: Sample J (Run 5 of 5) CONTROL - H_2O



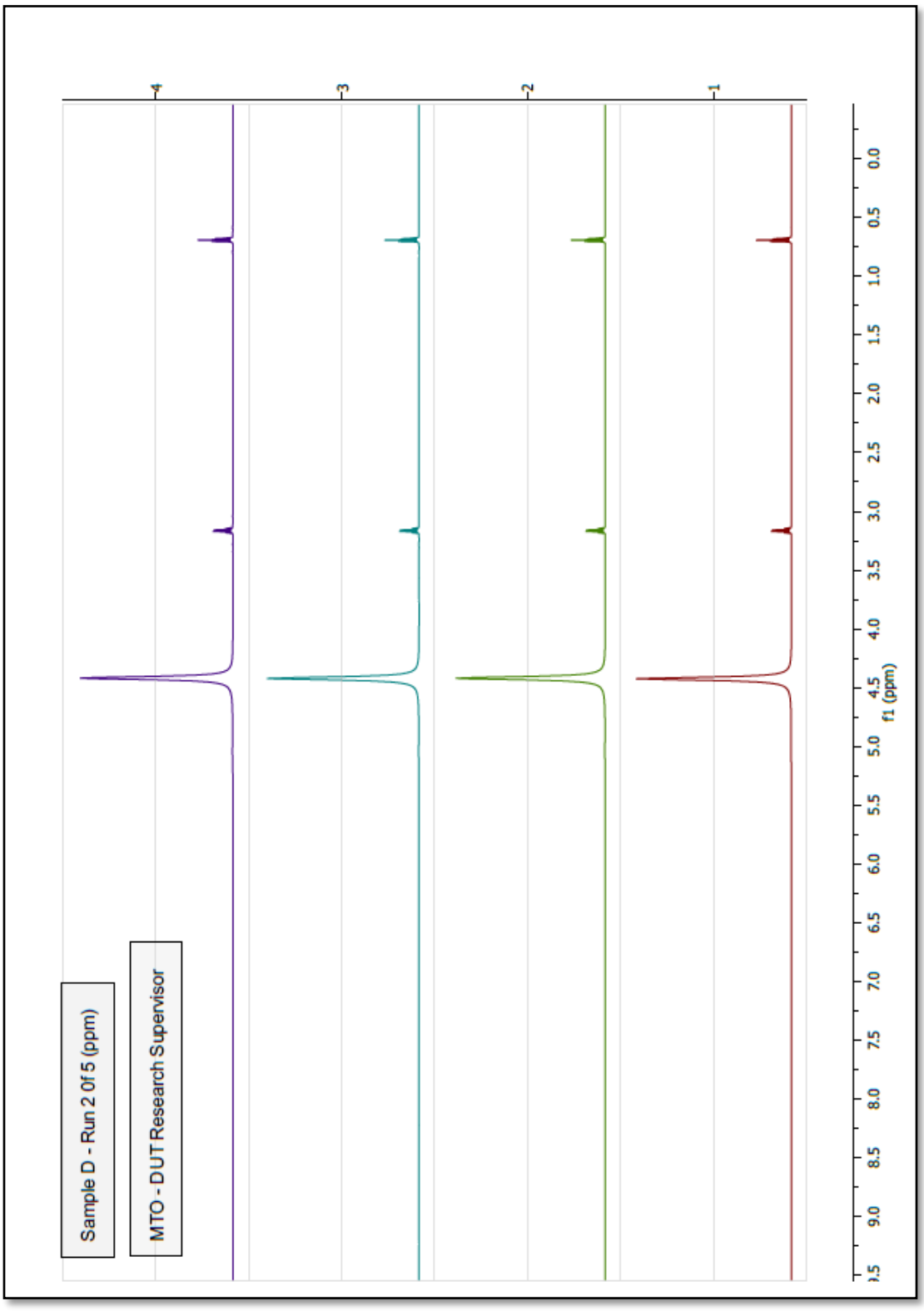
Graph 50: Sample A (Run 1-5 Stacked View) MTO - DUT Student Researcher



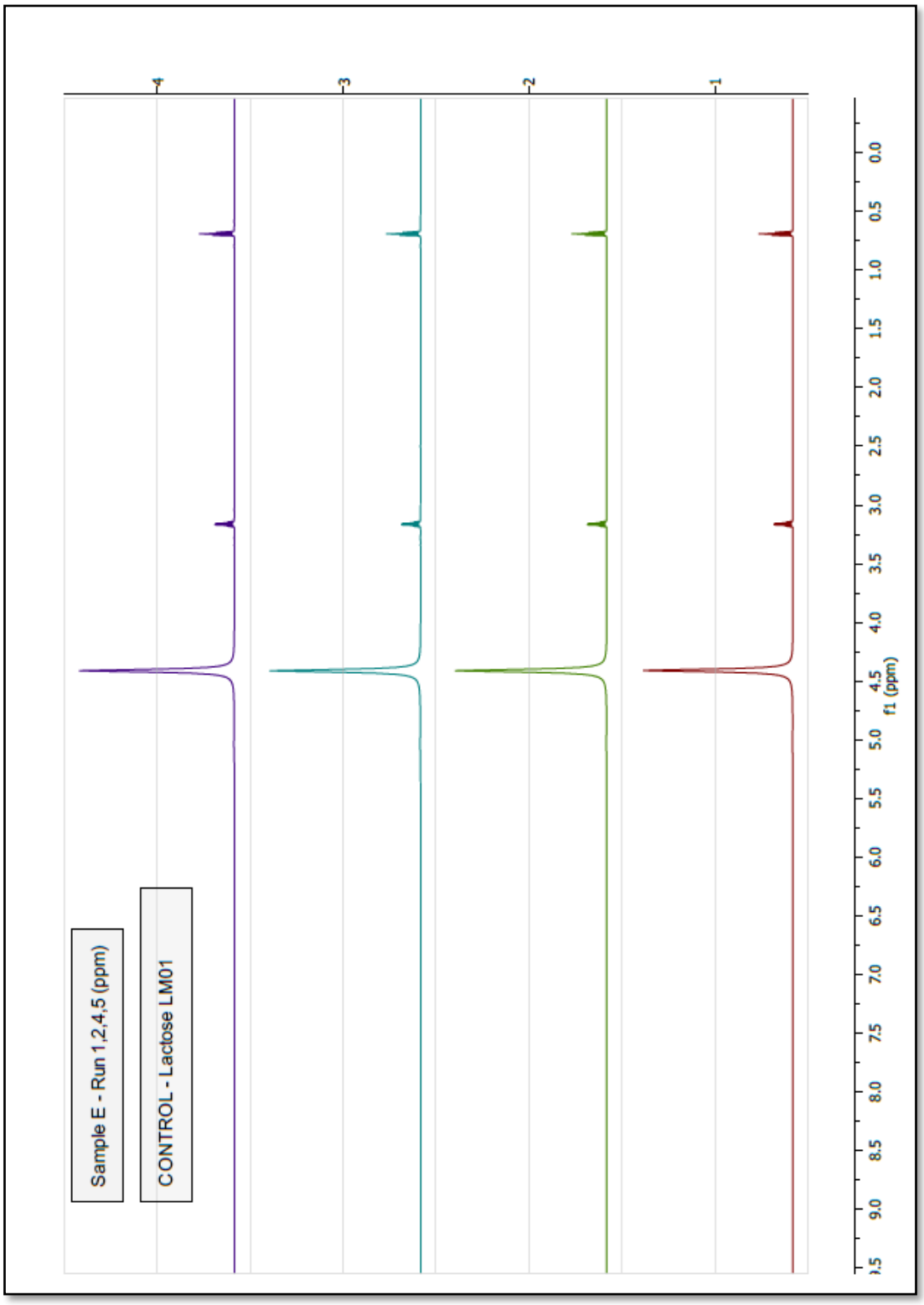
Graph 51: Sample B (Run 1-5 Stacked View) MTS - Roy & Co. (India)



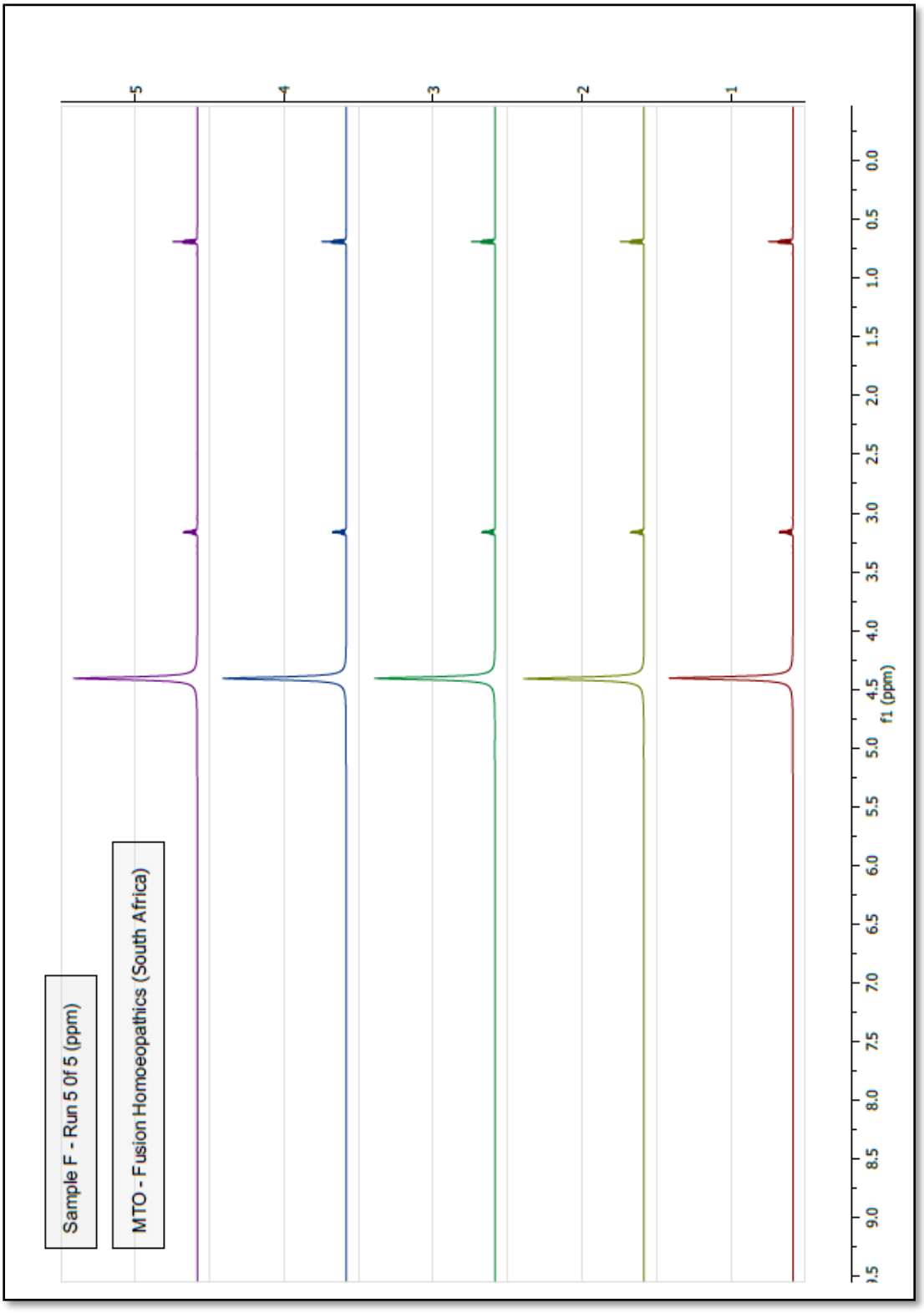
Graph 52: Sample A (Run 1-5 Stacked View) MTS -Helios (UK)



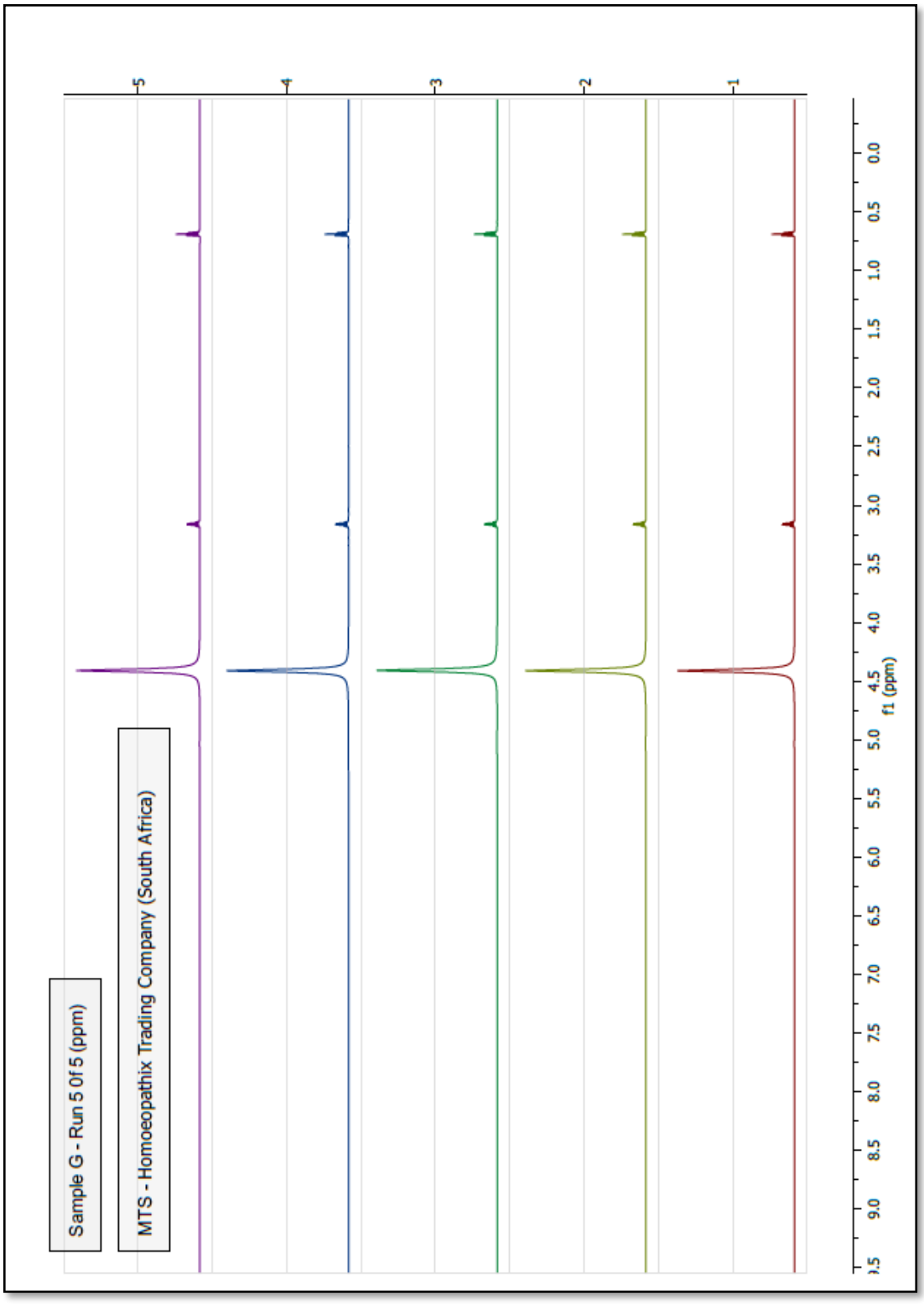
Graph 53: Sample D (Run 1-5 Stacked View) MTO - DUT Research Supervisor



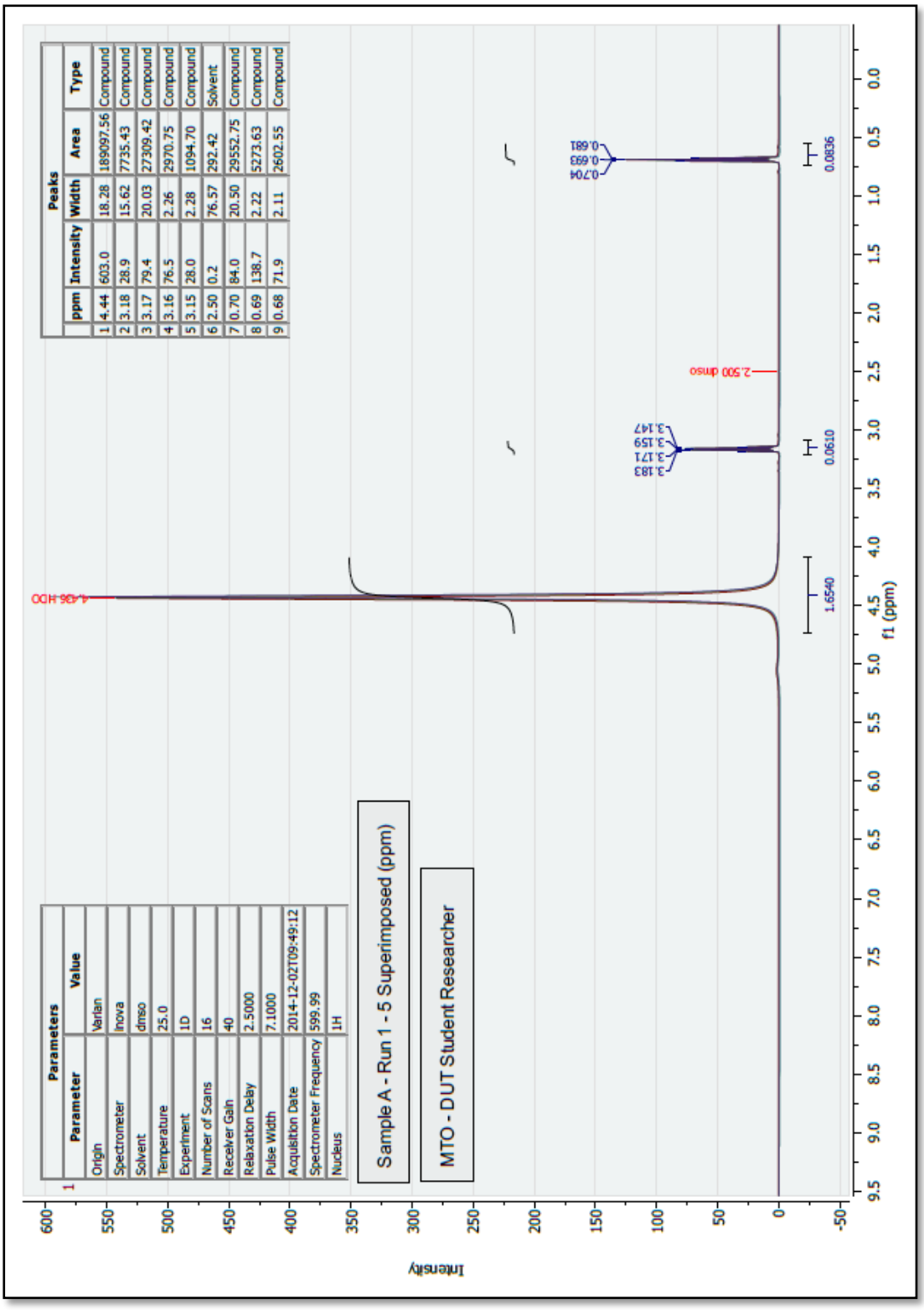
Graph 54: Sample A (Run 1,2,4,5 Stacked View) CONTROL - Lactose LM1



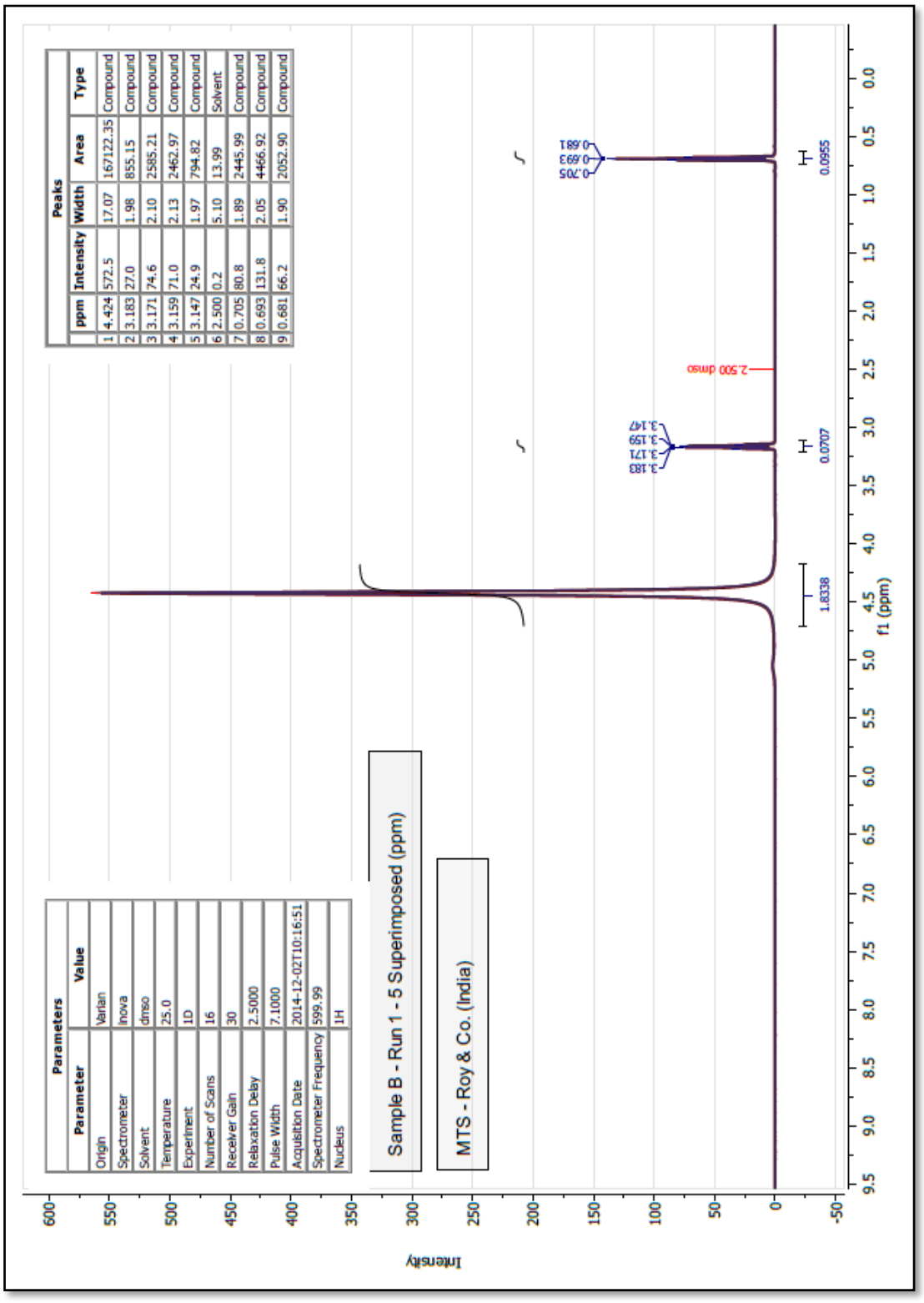
Graph 55: Sample F (Run 1-5 Stacked View) MTO - Fusion Homoeopathics (ZA)



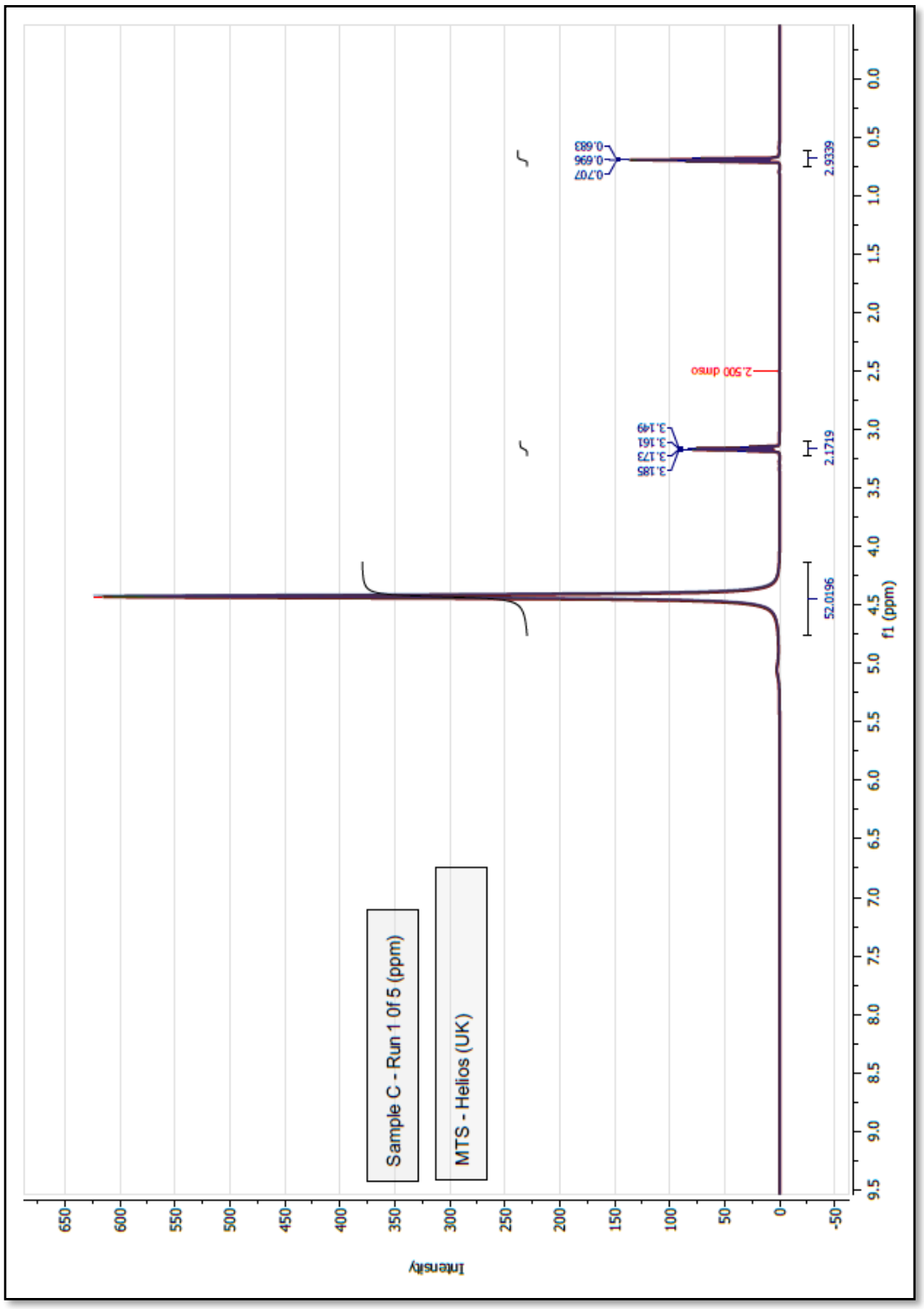
Graph 56: Sample G (Run 1-5 Stacked View) MTS - Homoeopathix Trading Co. (ZA)



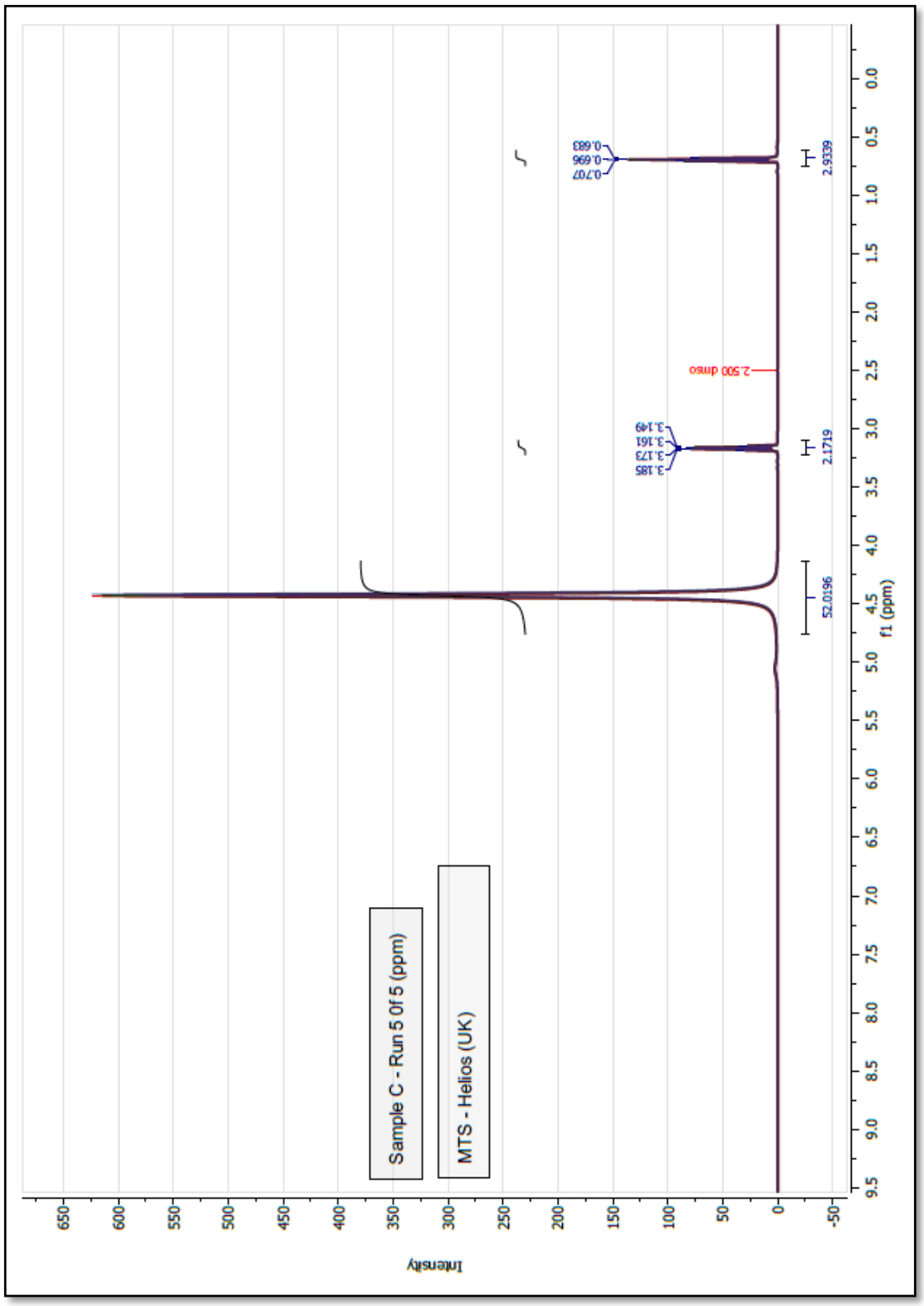
Graph 57: Sample A (Run 1-5 Superimposed) MTO - DUT Student Researcher



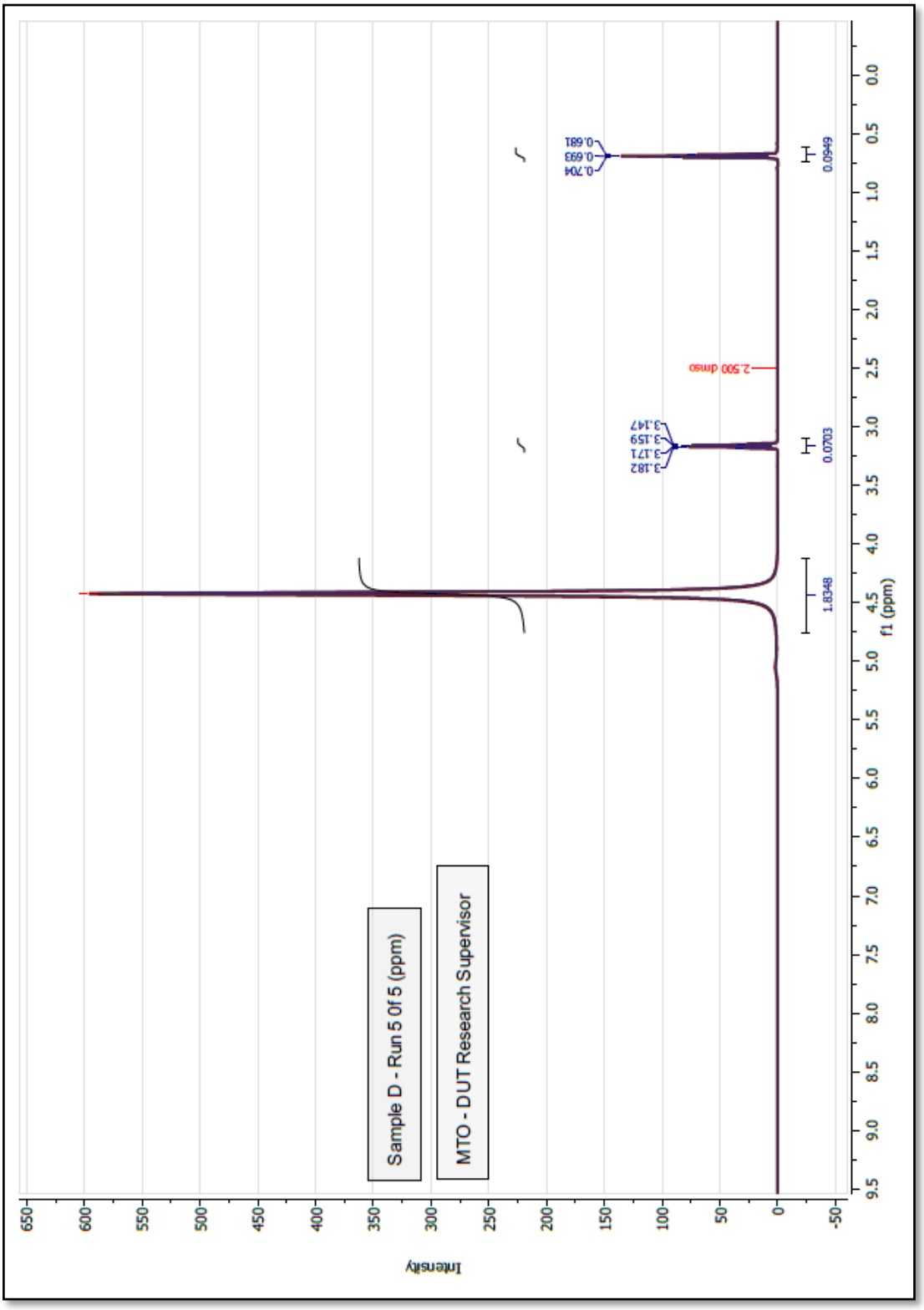
Graph 58: Sample B (Run 1-5 Superimposed) MTS - Roy & Co. (India)



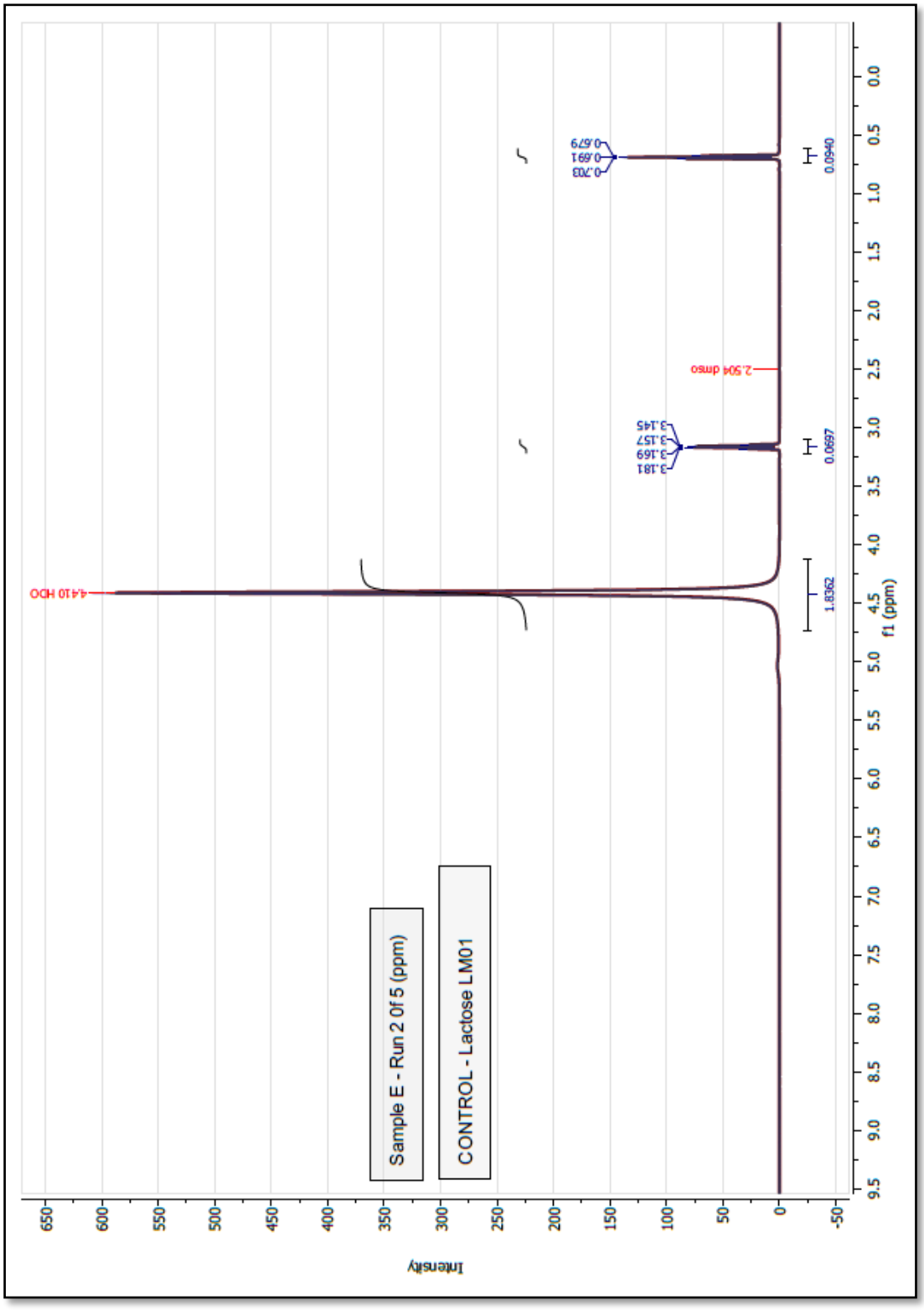
Graph 59: Sample C (Run 1-5 Superimposed) MTS - Helios (UK)



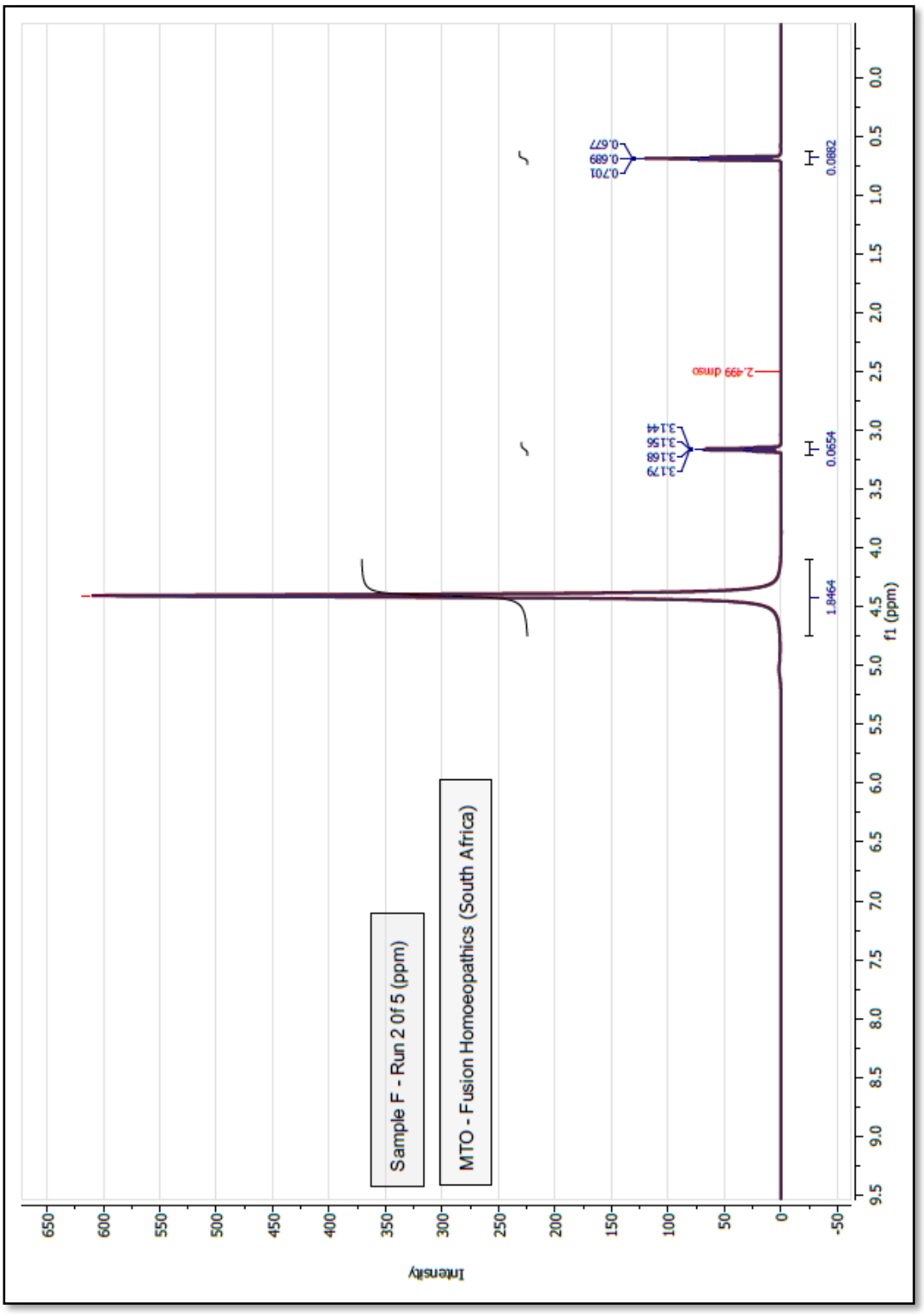
Graph 60: Sample C (Run 1-5 Superimposed) MTS - Helios (UK)



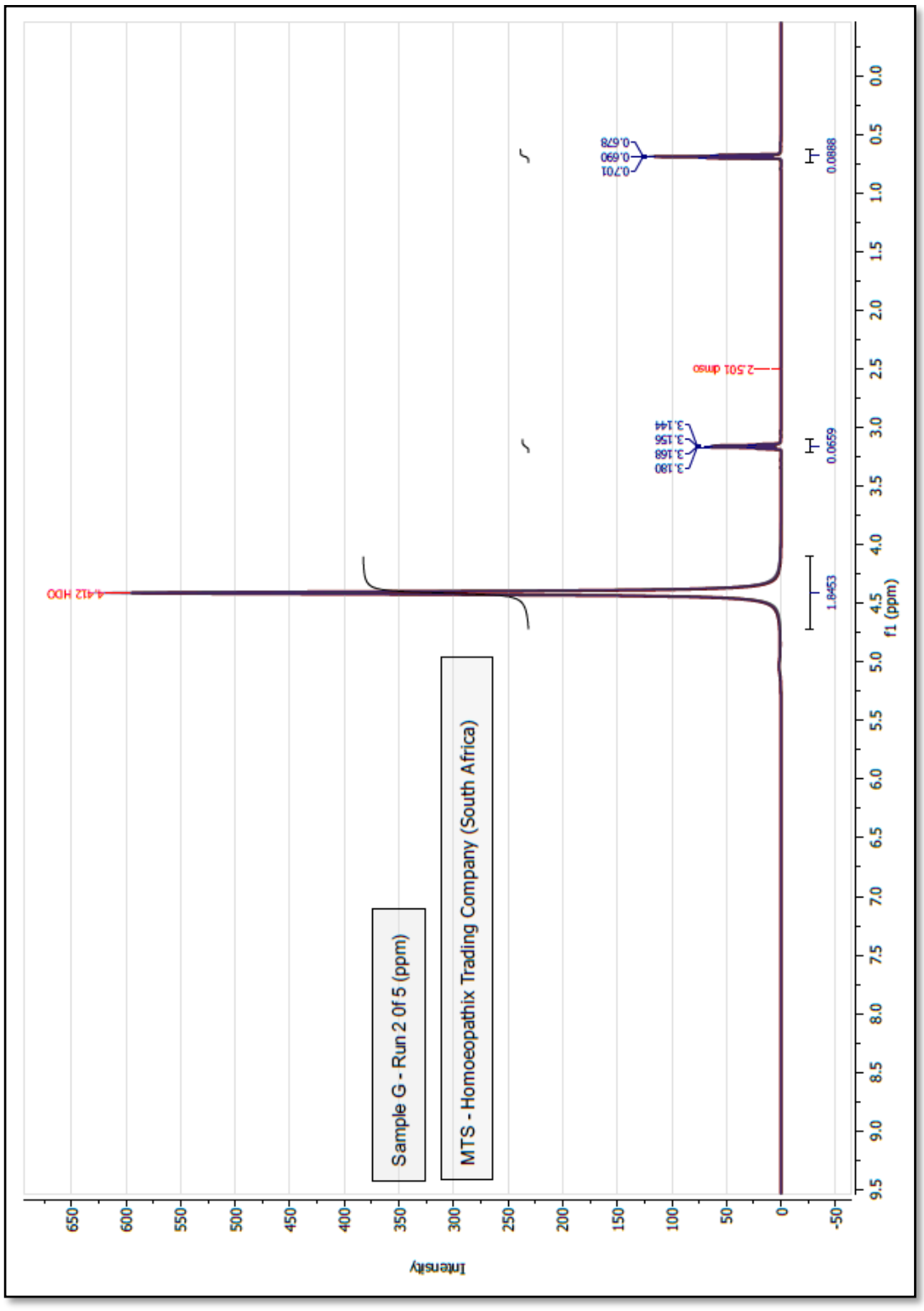
Graph 61: Sample D (Run 1-5 Superimposed) MTO - DUT Research Supervisor



Graph 62: Sample E (Run 1-5 Superimposed) CONTROL - Lactose LM1



Graph 63: Sample F (Run 1-5 Superimposed) MTO - Fusion Homoeopathics (ZA)



Graph 64: Sample G (Run 1-5 Superimposed) MTS - Homoeopathix Trading Co. (ZA)

Appendix F - Summary of data obtained from NMR spectroscopy

F.1 Chemical Shift values

	H_2O	CH_2	CH_3
Sample A - MTO - DUT Garth Miller			
Run 1	4,436	3,1650	0,6927
Run 2	4,428	3,1640	0,6920
Run 3	4,427	3,1640	0,6920
Run 4	4,423	3,1640	0,6917
Run 5	4,423	3,1640	0,6917
Sample B - MTS - Roy & Co.(India)			
Run 1	4,424	3,1650	0,6930
Run 2	4,420	3,1648	0,6923
Run 3	4,422	3,1648	0,6923
Run 4	4,419	3,1648	0,6920
Run 5	4,422	3,1648	0,6917

C

Sample C - MTS - Helios (UK)			
Run 1	4,430	3,1670	0,6953
Run 2	4,429	3,1670	0,6953
Run 3	4,427	3,1670	0,6950
Run 4	4,418	3,1665	0,6947
Run 5	4,427	3,1670	0,6950
Sample D - MTO - Izel Botha			
Run 1	4,425	3,1648	0,6927
Run 2	4,420	3,1643	0,6920
Run 3	4,422	3,1648	0,6923
Run 4	4,419	3,1640	0,6920
Run 5	4,419	3,1643	0,6920
Sample E - CONTROL - Lactose LM1			
Run 1	4,410	3,1630	0,6910
Run 2	4,412	3,1638	0,6910
Run 3	0,000	0,0000	0,0000
Run 4	4,413	3,1638	0,6913
Run 5	4,412	3,1638	0,6910
Sample F - MTO - Fusion Homoeopathics (South Africa)			
Run 1	4,405	3,1618	0,6890
Run 2	4,411	3,1620	0,6893
Run 3	4,407	3,1618	0,6890
Run 4	4,408	3,1618	0,6890
Run 5	4,408	3,1618	0,6890

Sample G - MTS - Homoeopathix Trading Co. (South Africa)			
Run 1	4,412	3,1620	0,6897
Run 2	4,411	3,1620	0,6893
Run 3	4,408	3,1618	0,6890
Run 4	4,411	3,1620	0,6897
Run 5	4,410	3,1618	0,6890

Sample H - CONTROL - Lactose			
Run 1	4,731	3,2618	0,5650
Run 2	4,731	3,2618	0,5650
Run 3	4,731	3,2618	0,5650
Run 4	4,731	3,2618	0,5650
Run 5	4,731	3,2618	0,5650

Sample I - CONTROL - 95% ROH			
Run 1	4,727	3,2600	0,5643
Run 2	4,727	3,2600	0,5643
Run 3	4,730	3,2630	0,5673
Run 4	4,727	3,2600	0,5643
Run 5	4,730	3,2630	0,5673

Sample J - CONTROL - H2O			
Run 1	4,727	3,2588	0,5623
Run 2	4,727	3,2588	0,5623
Run 3	4,728	3,2588	0,5623
Run 4	4,727	3,2588	0,5623
Run 5	4,728	3,2588	0,5623

F.2 Relative integration values

	H_2O	CH_2	CH_3
Sample A - MTO - DUT Garth Miller			
Run 1	1,654	0,061	0,084
Run 2	1,841	0,068	0,091
Run 3	1,851	0,063	0,085
Run 4	1,852	0,063	0,085
Run 5	1,839	0,068	0,093
Sample B - MTS - Roy & Co.(India)			
Run 1	1,834	0,071	0,096
Run 2	1,833	0,071	0,096
Run 3	1,833	0,071	0,096
Run 4	1,833	0,071	0,096
Run 5	1,833	0,071	0,096
Sample C - MTS - Helios (UK)			
Run 1	1,821	0,076	0,103
Run 2	1,820	0,076	0,103
Run 3	1,821	0,076	0,103
Run 4	1,821	0,076	0,103
Run 5	1,821	0,076	0,103
Sample D - MTO - Izel Botha			
Run 1	1,835	0,070	0,095
Run 2	1,834	0,071	0,095
Run 3	1,834	0,071	0,095
Run 4	1,833	0,071	0,096
Run 5	1,834	0,071	0,095
Sample E - CONTROL - Lactose LM1			
Run 1	1,836	0,070	0,094
Run 2	1,836	0,070	0,095
Run 3			
Run 4	1,836	0,070	0,094
Run 5	1,836	0,070	0,094
Sample F - MTO - Fusion Homoeopathics (South Africa)			
Run 1	1,846	0,065	0,088
Run 2	1,847	0,065	0,088
Run 3	1,846	0,066	0,089
Run 4	1,845	0,066	0,089
Run 5	1,846	0,066	0,089

Sample G - MTS - Homoeopathix Trading Co. (South Africa)			
Run 1	1,845	0,066	0,089
Run 2	1,845	0,066	0,089
Run 3	1,844	0,065	0,091
Run 4	1,845	0,066	0,089
Run 5	1,844	0,066	0,091

Problematic Data

Sample H - CONTROL - Lactose			
Run 1	*0,000*	*0,000*	*0,000*
Run 2	*0,000*	*0,000*	*0,000*
Run 3	*0,000*	*0,000*	*0,000*
Run 4	*0,000*	*0,000*	*0,000*
Run 5	*0,000*	*0,000*	*0,000*

Sample I - CONTROL - 95% ROH			
Run 1	*0,000*	*0,000*	*0,000*
Run 2	*0,000*	*0,000*	*0,000*
Run 3	*0,000*	*0,000*	*0,000*
Run 4	*0,000*	*0,000*	*0,000*
Run 5	*0,000*	*0,000*	*0,000*

Sample J - CONTROL - H2O			
Run 1	*0,000*	*0,000*	*0,000*
Run 2	*0,000*	*0,000*	*0,000*
Run 3	*0,000*	*0,000*	*0,000*
Run 4	*0,000*	*0,000*	*0,000*
Run 5	*0,000*	*0,000*	*0,000*

Appendix G - Results Obtained From Statistical Analysis

G.1 Kruskal-Wallis Chemical Shift

G.1.1 Statistical Shift

Descriptives Homogeneity		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Min.	Max.
						Lower Bound	Upper Bound		
H_2O	MTO Garth	5	4.43	.01	.00	4.42	4.43	4.423	4.436
	MTS Roy&Co.	5	4.42	.00	.00	4.42	4.42	4.419	4.424
	MTS Helios	5	4.43	.00	.00	4.42	4.43	4.418	4.430
	MTO Izel	5	4.42	.00	.00	4.42	4.42	4.419	4.425
	Control Lactose LM1	4	4.41	.00	.00	4.41	4.41	4.410	4.413
	MTO Fusion	5	4.41	.00	.00	4.41	4.41	4.405	4.411
	MTS Homoeopathix	5	4.41	.00	.00	4.41	4.41	4.408	4.412
	Total	***34***	***4.42***	***.01***	***.00***	***4.42***	***4.42***	***4.405***	***4.436***
CH_2	MTO Garth	5	3.16	.00	.00	3.16	3.16	3.164	3.165
	MTS Roy&Co.	5	3.16	.00	.00	3.16	3.16	3.165	3.165
	MTS Helios	5	3.17	.00	.00	3.17	3.17	3.167	3.167
	MTO Izel	5	3.16	.00	.00	3.16	3.16	3.164	3.165
	Control Lactose LM1	4	3.16	.00	.00	3.16	3.16	3.163	3.164
	MTO Fusion	5	3.16	.00	.00	3.16	3.16	3.162	3.162
	MTS Homoeopathix	5	3.16	.00	.00	3.16	3.16	3.162	3.162
	Total	***34***	**3.16**	***.00***	***.00***	**3.16**	**3.16**	***3.162***	***3.167***
CH_3	MTO Garth	5	.69	.00	.00	.69	.69	.692	.693
	MTS Roy&Co.	5	.69	.00	.00	.69	.69	.692	.693
	MTS Helios	5	.70	.00	.00	.69	.70	.695	.695
	MTO Izel	5	.69	.00	.00	.69	.69	.692	.693
	Control Lactose LM1	4	.69	.00	.00	.69	.69	.691	.691
	MTO Fusion	5	.69	.00	.00	.69	.69	.689	.689
	MTS Homoeopathix	5	.69	.00	.00	.69	.69	.689	.690
	Total	***34***	**.69**	***.00***	***.00***	**.69**	**.69**	***.689***	***.695***

G.1.2 Test of Homogeneity of Variances

	Levene Statistic	df1	df2	Sig.
H_2O	1.45	6	27	.232
CH_2	2.98	6	27	.023
CH_3	1.04	6	27	.424

G.1.3 Chemical Shift Analysis Of Variance (ANOVA)

		Sum of Squares	*df*	*Mean Square*	*df*	*Sig.*
H_2O	Between Groups	.00	6	.00	29.87	**.000**
	Within Groups	.00	27	.00		
	Total	.00	33			
CH_2	Between Groups	.00	6	.00	199.35	**.000**
	Within Groups	.00	27	.00		
	Total	.00	33			
CH_3	Between Groups	.00	6	.00	194.26	**.000**
	Within Groups	.00	27	.00		
	Total	.00	33			

G.2 Kruskal-Wallis Relative Integration

G.2.1 Statistical Shift

		N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Min.	Max.
						Lower Bound	Upper Bound		
H_2O	MTO Garth	5	.92	.00	.00	.92	.93	.919400	.925750
	MTS Roy&Co.	5	.92	.00	.00	.92	.92	.916350	.916900
	MTS Helios	5	.91	.00	.00	.91	.91	.910200	.910646
	MTO Izel	5	.92	.00	.00	.92	.92	.916700	.917400
	Control Lactose LM1	4	.92	.00	.00	.92	.92	.917800	.918146
	MTO Fusion	5	.92	.00	.00	.92	.92	.922746	.923350
	MTS Homoeopathix	5	.92	.00	.00	.92	.92	.921800	.922700
	Total	***34***	***.92***	***.00***	***.00***	***.92***	***.92***	***.910200***	***.925750***
CH_2	MTO Garth	5	.03	.00	.00	.03	.03	.031650	.033915
	MTS Roy&Co.	5	.04	.00	.00	.04	.04	.035350	.035550
	MTS Helios	5	.04	.00	.00	.04	.04	.038002	.038200
	MTO Izel	5	.04	.00	.00	.04	.04	.035150	.035450
	Control Lactose LM1	4	.03	.00	.00	.03	.03	.034850	.034950
	MTO Fusion	5	.03	.00	.00	.03	.03	.032600	.032898
	MTS Homoeopathix	5	.03	.00	.00	.03	.03	.032500	.032950
	Total	***34***	***.03***	***.00***	***.00***	***.03***	***.04***	***.031650***	***.038200***
CH_3	MTO Garth	5	.04	.00	.00	.04	.05	.042600	.046700
	MTS Roy&Co.	5	.05	.00	.00	.05	.05	.047750	.048100
	MTS Helios	5	.05	.00	.00	.05	.05	.051350	.051650
	MTO Izel	5	.05	.00	.00	.05	.05	.047450	.047850
	Control Lactose LM1	4	.05	.00	.00	.05	.05	.047002	.047250
	MTO Fusion	5	.04	.00	.00	.04	.04	.044050	.044402
	MTS Homoeopathix	5	.04	.00	.00	.04	.05	.044350	.045450
	Total	***34***	***.05***	***.00***	***.00***	***.05***	***.05***	***.042600***	***.051650***

G.2.2 Test of Homogeneity of Variances

	Levene Statistic	df1	df2	Sig.
H_2O	45.96	6	27	.000
CH_2	54.52	6	27	.000
CH_3	32.65	6	27	.000

G.2.3 Relative Integration Analysis Of Variance (ANOVA)

		Sum of Squares	*df*	*Mean Square*	*df*	*Sig.*
H_2O	Between Groups	.00	6	.00	62.93	.000
	Within Groups	.00	27	.00		
	Total	.00	33			
CH_2	Between Groups	.00	6	.00	83.58	.000
	Within Groups	.00	27	.00		
	Total	.00	33			
CH_3	Between Groups	.00	6	.00	48.38	.000
	Within Groups	.00	27	.00		
	Total	.00	33			

G.3 Mann-Whitney Chemical Shift

T-Test

Missing = Analysis

Criteria = CI(0.95).

G.3.1 Control Lactose LM1 comparison to MTO Garth

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTO Garth	5	4.43	.01	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTO Garth	5	3.16	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTO Garth	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	2.59	.151	-5.68	7.00	.001	-.02	.00	-.02	-.01
	Not assumed			-6.36	4.55	.002	-.02	.00	-.02	-.01
CH_2	Assumed	.02	.905	-2.09	7.00	.075	.00	.00	.00	.00
	Not assumed			-2.12	6.86	.072	.00	.00	.00	.00
CH_3	Assumed	1.26	.299	-4.35	7.00	.003	.00	.00	.00	.00
	Not assumed			-4.78	5.26	.004	.00	.00	.00	.00

G.3.2 Control Lactose LM1 comparison to MTS Roy & Co.

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTS Roy & Co.	5	4.42	.00	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTS Roy & Co.	5	3.16	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTS Roy & Co.	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.21	.307	-8.52	7.00	.000	-.01	.00	-.01	-.01
	Not assumed			-8.98	6.79	.000	-.01	.00	-.01	-.01
CH_2	Assumed	6.59	.037	-6.83	7.00	.000	.00	.00	.00	.00
	Not assumed			-6.08	3.24	.007	.00	.00	.00	.00
CH_3	Assumed	1.76	.227	-4.67	7.00	.002	.00	.00	.00	.00
	Not assumed			-5.19	4.93	.004	.00	.00	.00	.00

G.3.3 Control Lactose LM1 comparison to MTS Helios

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTS Helios	5	4.43	.00	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTS Helios	5	3.17	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTS Helios	5	.70	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	2.33	.171	-5.83	7.00	.001	-.01	.00	-.02	-.01
	Not assumed			-6.50	4.68	.002	-.01	.00	-.02	-.01
CH_2	Assumed	1.59	.248	-15.78	7.00	.000	.00	.00	.00	.00
	Not assumed			-14.76	4.48	.000	.00	.00	.00	.00
CH_3	Assumed	1.16	.317	-27.81	7.00	.000	.00	.00	.00	.00
	Not assumed			-29.52	6.61	.000	.00	.00	.00	.00

G.3.4 Control Lactose LM1 comparison to MTO Izel Botha

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTO Izel	5	4.42	.00	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTO Izel	5	3.16	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTO Izel	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	2.56	.154	-6.58	7.00	.000	-.01	.00	-.01	-.01
	Not assumed			-7.10	6.06	.000	-.01	.00	-.01	-.01
CH_2	Assumed	.01	.918	-3.36	7.00	.012	.00	.00	.00	.00
	Not assumed			-3.30	6.10	.016	.00	.00	.00	.00
CH_3	Assumed	2.32	.171	-6.63	7.00	.000	.00	.00	.00	.00
	Not assumed			-7.17	6.02	.000	.00	.00	.00	.00

G.3.5 Control Lactose LM1 comparison to MTO Fusion Homoeopathics

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTO Fusion Homoeopathics	5	4.41	.00	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTO Fusion Homoeopathics	5	3.16	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTO Fusion Homoeopathics	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	F	Sig.	t	df	Sig. (2-tailed)	Mean Diff.	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
H_2O	Assumed	.49	.506	3.21	7.00	.015	.00	.00	.00	.01
	Not assumed			3.42	6.53	.012	.00	.00	.00	.01
CH_2	Assumed	6.59	.037	9.70	7.00	.000	.00	.00	.00	.00
	Not assumed			8.63	3.24	.002	.00	.00	.00	.00
CH_3	Assumed	.10	.762	21.28	7.00	.000	.00	.00	.00	.00
	Not assumed			20.98	6.17	.000	.00	.00	.00	.00

G.3.6 Control Lactose LM1 comparison to MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	Control Lactose LM1	4	4.41	.00	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	Control Lactose LM1	4	3.16	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	Control Lactose LM1	4	.69	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.20	.666	1.43	7.00	.197	.00	.00	.00	.00
	Not assumed			1.46	6.97	.188	.00	.00	.00	.00
CH_2	Assumed	5.31	.055	9.12	7.00	.000	.00	.00	.00	.00
	Not assumed			8.16	3.36	.003	.00	.00	.00	.00
CH_3	Assumed	5.09	.059	9.15	7.00	.000	.00	.00	.00	.00
	Not assumed			9.98	5.64	.000	.00	.00	.00	.00

G.3.7 MTO Garth Comparison to MTS Roy & Co.

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Garth	5	4.43	.01	.00
	MTS Roy & Co.	5	4.42	.00	.00
CH_2	MTO Garth	5	3.16	.00	.00
	MTS Roy & Co.	5	3.16	.00	.00
CH_3	MTO Garth	5	.69	.00	.00
	MTS Roy & Co.	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*T*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.90	.205	2.37	8.00	.045	.01	.00	.00	.01
	Not assumed			2.37	5.06	.064	.01	.00	.00	.01
CH_2	Assumed	4.38	.070	-3.14	8.00	.014	.00	.00	.00	.00
	Not assumed			-3.14	4.32	.031	.00	.00	.00	.00
CH_3	Assumed	.09	.771	-.85	8.00	.421	.00	.00	.00	.00
	Not assumed			-.85	7.79	.421	.00	.00	.00	.00

G.3.8 MTO Garth Comparison to MTS Helios

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	4.43	.01	.00
	MTS Helios	5	4.43	.00	.00
CH_2	MTO Garth	5	3.16	.00	.00
	MTS Helios	5	3.17	.00	.00
CH_3	MTO Garth	5	.69	.00	.00
	MTS Helios	5	.70	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	F	Sig.	t	df	Sig. (2-tailed)	Mean Diff.	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
H_2O	Assumed	.04	.849	.38	8.00	.717	.00	.00	-.01	.01
	Not assumed			.38	7.90	.717	.00	.00	-.01	.01
CH_2	Assumed	1.42	.267	-12.07	8.00	.000	.00	.00	.00	.00
	Not assumed			-12.07	5.88	.000	.00	.00	.00	.00
CH_3	Assumed	.35	.570	-14.17	8.00	.000	.00	.00	.00	.00
	Not assumed			-14.17	6.64	.000	.00	.00	.00	.00

G.3.9 MTO Garth Comparison to MTO Izel

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	4.43	.01	.00
	MTO Izel	5	4.42	.00	.00
CH_2	MTO Garth	5	3.16	.00	.00
	MTO Izel	5	3.16	.00	.00
CH_3	MTO Garth	5	.69	.00	.00
	MTO Izel	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	F	Sig.	T	df	Sig. (2-tailed)	Mean Diff.	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
H_2O	Assumed	1.10	.326	2.43	8.00	.041	.01	.00	.00	.01
	Not assumed			2.43	5.75	.053	.01	.00	.00	.01
CH_2	Assumed	.06	.819	-.94	8.00	.373	.00	.00	.00	.00
	Not assumed			-.94	7.57	.374	.00	.00	.00	.00
CH_3	Assumed	.05	.825	-.79	8.00	.454	.00	.00	.00	.00
	Not assumed			-.79	7.44	.456	.00	.00	.00	.00

G.3.10 MTO Garth Comparison to MTO Fusion Homoeopathics

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Garth	5	4.43	.01	.00
	MTO Fusion Homoeopathics	5	4.41	.00	.00
CH_2	MTO Garth	5	3.16	.00	.00
	MTO Fusion Homoeopathics	5	3.16	.00	.00
CH_3	MTO Garth	5	.69	.00	.00
	MTO Fusion Homoeopathics	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.86	.210	7.63	8.00	.000	.02	.00	.01	.03
	Not assumed			7.63	5.29	.000	.02	.00	.01	.03
CH_2	Assumed	4.38	.070	11.57	8.00	.000	.00	.00	.00	.00
	Not assumed			11.57	4.32	.000	.00	.00	.00	.00
CH_3	Assumed	1.91	.204	15.39	8.00	.000	.00	.00	.00	.00
	Not assumed			15.39	4.85	.000	.00	.00	.00	.00

G.3.11 MTO Garth Comparison to MTS Homoeopathix

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	4.43	.01	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	MTO Garth	5	3.16	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	MTO Garth	5	.69	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	F	Sig.	t	df	Sig. (2-tailed)	Mean Diff.	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
H_2O	Assumed	2.71	.139	6.87	8.00	.000	.02	.00	.01	.02
	Not assumed			6.87	4.65	.001	.02	.00	.01	.02
CH_2	Assumed	3.46	.100	11.07	8.00	.000	.00	.00	.00	.00
	Not assumed			11.07	4.48	.000	.00	.00	.00	.00
CH_3	Assumed	.01	.910	11.13	8.00	.000	.00	.00	.00	.00
	Not assumed			11.13	7.82	.000	.00	.00	.00	.00

G.3.12 MTS Roy & Co. to MTS Helios

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	4.42	.00	.00
	MTS Helios	5	4.43	.00	.00
CH_2	MTS Roy & Co.	5	3.16	.00	.00
	MTS Helios	5	3.17	.00	.00
CH_3	MTS Roy & Co.	5	.69	.00	.00
	MTS Helios	5	.70	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.52	.252	-2.08	8.00	.071	.00	.00	-.01	.00
	Not assumed			-2.08	5.30	.088	.00	.00	-.01	.00
CH_2	Assumed	2.21	.176	-19.13	8.00	.000	.00	.00	.00	.00
	Not assumed			-19.13	5.25	.000	.00	.00	.00	.00
CH_3	Assumed	.80	.397	-11.51	8.00	.000	.00	.00	.00	.00
	Not assumed			-11.51	6.02	.000	.00	.00	.00	.00

G.3.13 MTS Roy & Co. to MTO Izel

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	4.42	.00	.00
	MTO Izel	5	4.42	.00	.00
CH_2	MTS Roy & Co.	5	3.16	.00	.00
	MTO Izel	5	3.16	.00	.00
CH_3	MTS Roy & Co.	5	.69	.00	.00
	MTO Izel	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*T*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.48	.509	.28	8.00	.788	.00	.00	.00	.00
	Not assumed			.28	7.49	.788	.00	.00	.00	.00
CH_2	Assumed	11.30	.010	2.47	8.00	.039	.00	.00	.00	.00
	Not assumed			2.47	4.52	.062	.00	.00	.00	.00
CH_3	Assumed	.32	.588	.23	8.00	.821	.00	.00	.00	.00
	Not assumed			.23	6.80	.822	.00	.00	.00	.00

G.3.14 MTS Roy & Co. to MTO Fusion Homoeopathics

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	4.42	.00	.00
	MTO Fusion Homoeopathics	5	4.41	.00	.00
CH_2	MTS Roy & Co.	5	3.16	.00	.00
	MTO Fusion Homoeopathics	5	3.16	.00	.00
CH_3	MTS Roy & Co.	5	.69	.00	.00
	MTO Fusion Homoeopathics	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.01	.921	10.43	8.00	.000	.01	.00	.01	.02
	Not assumed			10.43	7.91	.000	.01	.00	.01	.02
CH_2	Assumed	.00	1.000	53.03	8.00	.000	.00	.00	.00	.00
	Not assumed			53.03	8.00	.000	.00	.00	.00	.00
CH_3	Assumed	2.56	.148	14.28	8.00	.000	.00	.00	.00	.00
	Not assumed			14.28	4.61	.000	.00	.00	.00	.00

G.3.15 MTS Roy & Co. to MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	4.42	.00	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	MTS Roy & Co.	5	3.16	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	MTS Roy & Co.	5	.69	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.49	.505	9.96	8.00	.000	.01	.00	.01	.01
	Not assumed			9.96	7.54	.000	.01	.00	.01	.01
CH_2	Assumed	1.52	.252	46.17	8.00	.000	.00	.00	.00	.00
	Not assumed			46.17	7.69	.000	.00	.00	.00	.00
CH_3	Assumed	.07	.801	10.94	8.00	.000	.00	.00	.00	.00
	Not assumed			10.94	7.30	.000	.00	.00	.00	.00

G.3.16 MTS Helios Comparison to MTO Izel

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	4.43	.00	.00
	MTO Izel	5	4.42	.00	.00
CH_2	MTS Helios	5	3.17	.00	.00
	MTO Izel	5	3.16	.00	.00
CH_3	MTS Helios	5	.70	.00	.00
	MTO Izel	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.76	.408	2.15	8.00	.064	.01	.00	.00	.01
	Not assumed			2.15	6.12	.074	.01	.00	.00	.01
CH_2	Assumed	2.20	.177	13.23	8.00	.000	.00	.00	.00	.00
	Not assumed			13.23	6.79	.000	.00	.00	.00	.00
CH_3	Assumed	.29	.606	16.09	8.00	.000	.00	.00	.00	.00
	Not assumed			16.09	7.68	.000	.00	.00	.00	.00

G.3.17 MTS Helios Comparison to MTO Fusion

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	4.43	.00	.00
	MTO Fusion Homoeopathics	5	4.41	.00	.00
CH_2	MTS Helios	5	3.17	.00	.00
	MTO Fusion Homoeopathics	5	3.16	.00	.00
CH_3	MTS Helios	5	.70	.00	.00
	MTO Fusion Homoeopathics	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.49	.257	7.86	8.00	.000	.02	.00	.01	.02
	Not assumed			7.86	5.59	.000	.02	.00	.01	.02
CH_2	Assumed	2.21	.176	46.98	8.00	.000	.01	.00	.00	.01
	Not assumed			46.98	5.25	.000	.01	.00	.00	.01
CH_3	Assumed	1.97	.198	47.14	8.00	.000	.01	.00	.01	.01
	Not assumed			47.14	6.11	.000	.01	.00	.01	.01

G.3.18 MTS Helios Comparison to MTS

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	4.43	.00	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	MTS Helios	5	3.17	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	MTS Helios	5	.70	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*T*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	2.34	.165	7.07	8.00	.000	.02	.00	.01	.02
	Not assumed			7.07	4.80	.001	.02	.00	.01	.02
CH_2	Assumed	1.11	.323	44.72	8.00	.000	.00	.00	.00	.00
	Not assumed			44.72	5.82	.000	.00	.00	.00	.00
CH_3	Assumed	1.27	.292	29.66	8.00	.000	.01	.00	.01	.01
	Not assumed			29.66	7.25	.000	.01	.00	.01	.01

G.3.19 MTO Izel Comparison to MTO Fusion

Group Statistics

	Method	*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Izel	5	4.42	.00	.00
	MTO Fusion Homoeopathics	5	4.41	.00	.00
CH_2	MTO Izel	5	3.16	.00	.00
	MTO Fusion Homoeopathics	5	3.16	.00	.00
CH_3	MTO Izel	5	.69	.00	.00
	MTO Fusion Homoeopathics	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.43	.528	8.82	8.00	.000	.01	.00	.01	.02
	Not assumed			8.82	7.80	.000	.01	.00	.01	.02
CH_2	Assumed	11.30	.010	16.06	8.00	.000	.00	.00	.00	.00
	Not assumed			16.06	4.52	.000	.00	.00	.00	.00
CH_3	Assumed	3.52	.098	20.89	8.00	.000	.00	.00	.00	.00
	Not assumed			20.89	5.46	.000	.00	.00	.00	.00

G.3.20 MTO Izel Comparison to MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Izel	5	4.42	.00	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	MTO Izel	5	3.16	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	MTO Izel	5	.69	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
2	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.73	.224	7.99	8.00	.000	.01	.00	.01	.01
	Not assumed			7.99	6.52	.000	.01	.00	.01	.01
CH_2	Assumed	9.31	.016	15.34	8.00	.000	.00	.00	.00	.00
	Not assumed			15.34	4.77	.000	.00	.00	.00	.00
CH_3	Assumed	.27	.616	13.70	8.00	.000	.00	.00	.00	.00
	Not assumed			13.70	7.87	.000	.00	.00	.00	.00

G.3.21 MTS Fusion Homoeopathics Comparison to MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Fusion Homoeopathics	5	4.41	.00	.00
	MTS Homoeopathix	5	4.41	.00	.00
CH_2	MTO Fusion Homoeopathics	5	3.16	.00	.00
	MTS Homoeopathix	5	3.16	.00	.00
CH_3	MTO Fusion Homoeopathics	5	.69	.00	.00
	MTS Homoeopathix	5	.69	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.18	.682	-2.20	8.00	.059	.00	.00	-.01	.00
	Not assumed			-2.20	7.16	.063	.00	.00	-.01	.00
CH_2	Assumed	1.52	.252	-1.26	8.00	.242	.00	.00	.00	.00
	Not assumed			-1.26	7.69	.243	.00	.00	.00	.00
CH_3	Assumed	7.14	.028	-1.67	8.00	.134	.00	.00	.00	.00
	Not assumed			-1.67	5.15	.155	.00	.00	.00	.00

G.4 Mann Whitney Relative Integration

G.4.1 Control Lactose LM1 and MTO Garth

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Garth	5	.92	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	Control Lactose LM1	4	.03	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	Control Lactose LM1	4	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*T*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	44.35	.000	2.58	7.00	.036	.00	.00	.00	.01
	Not assumed			2.93	4.02	**.043**	.00	.00	.00	.01
CH_2	Assumed	66.55	.000	-3.10	7.00	.017	.00	.00	.00	.00
	Not assumed			-.352	4.02	**.024**	.00	.00	.00	.00
CH_3	Assumed	31.19	.001	-2.25	7.00	.059	.00	.00	.00	.00
	Not assumed			-2.55	4.03	**.063**	.00	.00	.00	.00

G.4.2 MTS Roy & Co. and Control Lactose LM1

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	.92	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTS Roy & Co.	5	.04	.00	.00
	Control Lactose LM1	4	.03	.00	.00
CH_3	MTS Roy & Co.	5	.05	.00	.00
	Control Lactose LM1	4	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.48	.512	-11.32	7.00	.000	.00	.00	.00	.00
	Not assumed			-11.81	6.94	**.000**	.00	.00	.00	.00
CH_2	Assumed	2.96	.129	11.86	7.00	.000	.00	.00	.00	.00
	Not assumed			12.63	6.51	**.000**	.00	.00	.00	.00
CH_3	Assumed	.10	.760	10.49	7.00	.000	.00	.00	.00	.00
	Not assumed			10.76	6.98	**.000**	.00	.00	.00	.00

G.4.3 MTS Helios and Control Lactose LM1

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	.91	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTS Helios	5	.04	.00	.00
	Control Lactose LM1	4	03	.00	.00
CH_3	MTS Helios	5	.05	.00	.00
	Control Lactose LM1	4	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.17	.695	-71.81	7.00	.000	-.01	.00	-.01	-.01
	Not assumed			-73.34	6.95	.000	-.01	.00	-.01	-.01
CH_2	Assumed	.30	.603	74.91	7.00	.000	.00	.00	.00	.00
	Not assumed			79.07	675	.000	.00	.00	.00	.00
CH_3	Assumed	.75	.414	54.66	7.00	.000	.00	.00	.00	.00
	Not assumed			56.30	7.00	.000	.00	.00	.00	.00

G.4.4 MTO Izel Botha and Control Lactose LM1

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Izel	5	.92	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTO Izel	5	.04	.00	.00
	Control Lactose LM1	4	.03	.00	.00
CH_3	MTO Izel	5	.05	.00	.00
	Control Lactose LM1	4	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	F	Sig.	t	df	Sig. (2-tailed)	Mean Diff.	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
H_2O	Assumed	.93	.367	-6.48	7.00	.000	.00	.00	.00	.00
	Not assumed			-6.93	6.39	.000	.00	.00	.00	.00
CH_2	Assumed	1.54	.255	6.83	7.00	.000	.00	.00	.00	.00
	Not assumed			7.45	5.62	.000	.00	.00	.00	.00
CH_3	Assumed	.33	.582	6.21	7.00	.000	.00	.00	.00	.00
	Not assumed			6.48	6.94	.000	.00	.00	.00	.00

G.4.5 MTO Fusion Homoeopathics and Control Lactose LM1

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Fusion Homoeopathics	5	.92	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTO Fusion Homoeopathics	5	.03	.00	.00
	Control Lactose LM1	4	.03	.00	.00
CH_3	MTO Fusion Homoeopathics	5	.04	.00	.00
	Control Lactose LM1	4	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	4.13	.082	34.16	7.00	.000	.01	.00	.00	.01
	Not assumed			36.59	6.34	.000	.01	.00	.00	.01
CH_2	Assumed	6.89	.034	-31.60	7.00	.000	.00	.00	.00	.00
	Not assumed			-34.72	5.33	.000	.00	.00	.00	.00
CH_3	Assumed	1.19	.311	-33.69	7.00	.000	.00	.00	.00	.00
	Not assumed			-35.13	6.95	.000	.00	.00	.00	.00

G.4.6 MTS Homoeopathix and Control Lactose LM1

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Homoeopathix	5	.92	.00	.00
	Control Lactose LM1	4	.92	.00	.00
CH_2	MTS Homoeopathix	5	.03	.00	.00
	Control Lactose LM1	4	.03	.00	.00
CH_3	MTS Homoeopathix	5	.05	.00	.00
	Control Lactose LM1	4	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*Df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	12.62	.009	-19.92	7.00	.000	.00	.00	.00	.00
	Not assumed			-22.00	5.11	.000	.00	.00	.00	.00
CH_2	Assumed	2.73	.142	20.60	7.00	.000	.00	.00	.00	.00
	Not assumed			23.03	4.60	.000	.00	.00	.00	.00
CH_3	Assumed	33.31	.001	8.40	7.00	.000	.00	.00	.00	.00
	Not assumed			9.45	4.36	.000	.00	.00	.00	.00

G.4.7 MTO Garth and MTS Roy & Co.

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	.92	.01	.00
	MTS Roy & Co.	5	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	MTS Roy & Co.	5	.04	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	MTS Roy & Co.	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	54.17	.000	3.87	8.00	.005	.01	.00	.00	.01
	Not assumed			3.87	4.03	.018	.01	.00	.00	.01
CH_2	Assumed	79.55	.000	-4.54	8.00	.002	.00	.00	.00	.00
	Not assumed		.	-4.54	4.04	.010	.00	.00	.00	.00
CH_3	Assumed	38.92	.000	-3.45	8.00	.009	.00	..	-.01	.00
	Not assumed			-3.45	4.03	.026	.00	.00	-.01	.00

G.4.8 MTO Garth and MTS Helios

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	.92	.01	.00
	MTS Helios	5	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	MTS Helios	5	.04	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	MTS Helios	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	55.56	.000	8.19	8.00	.000	.01	.00	.01	.02
	Not assumed			8.19	4.02	.001	.01	.00	.01	.02
CH_2	Assumed	80.52	.000	-9.52	8.00	.000	-.01	.00	-.01	.00
	Not assumed			-9.52	4.03	.001	-.01	.00	-.01	.00
CH_3	Assumed	38.40	.000	-7.34	8.00	.000	-.01	.00	-.01	.00
	Not assumed			-7.34	4.03	.002	-.01	.00	-.01	.00

G.4.9 MTO Garth and MTO Izel

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	.92	.00	.00
	MTO Izel	5	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	MTO Izel	5	.04	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	MTO Izel	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	51.84	.000	3.57	8.00	.007	.01	.00	.00	.01
	Not assumed			3.57	4.05	.023	.01	.00	.00	.01
CH_2	Assumed	73.43	.000	-4.27	8.00	.003	.00	.00	.00	.00
	Not assumed			-4.27	4.07	.013	.00	.00	.00	.00
CH_3	Assumed	37.99	.000	-3.14	8.00	.014	.00	.00	.00	.00
	Not assumed			-3.14	4.04	.034	.00	.00	-.01	.00

G.4.10 MTO Garth and MTO Fusion Homoeopathics

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Garth	5	.92	.00	.00
	MTO Fusion Homoeopathics	5	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	MTO Fusion Homoeopathics	5	.03	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	MTO Fusion Homoeopathics	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	51.65	.000	-.55	8.00	.599	.00	.00	.00	.00
	Not assumed			-.55	4.05	.613	.00	.00	.00	.00
CH_2	Assumed	72.46	.000	.39	8.00	.704	.00	.00	.00	.00
	Not assumed			.39	4.09	.713	.00	.00	.00	.00
CH_3	Assumed	37.77	.000	.63	8.00	.544	.00	.00	.00	.00
	Not assumed			.63	4.04	.560	.00	.00	.00	.00

G.4.11 MTO Garth and MTS Homoeopathix

Group Statistics

Method		N	Mean	Std. Deviation	S.E. Mean
H_2O	MTO Garth	5	.92	.00	.00
	MTS Homoeopathix	5	.92	.00	.00
CH_2	MTO Garth	5	.03	.00	.00
	MTS Homoeopathix	5	.03	.00	.00
CH_3	MTO Garth	5	.04	.00	.00
	MTS Homoeopathix	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	45.85	.000	-.11	8.00	.915	.00	.00	.00	.00
	Not assumed			-.11	4.13	.918	.00	.00	.00	.00
CH_2	Assumed	57.14	.000	.28	8.00	.786	.00	.00	.00	.00
	Not assumed			.28	4.20	.792	.00	.00	.00	.00
CH_3	Assumed	22.40	.000	.01	8.00	.995	.00	.00	.00	.00
	Not assumed			.01	4.56	.995	.00	.00	.00	.00

G.4.12 MTS Roy & Co. and MTS Helios

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	.92	.00	.00
	MTS Helios	5	.91	.00	.00
CH_2	MTS Roy & Co.	5	.04	.00	.00
	MTS Helios	5	.04	.00	.00
CH_3	MTS Roy & Co.	5	.05	.00	.00
	MTS Helios	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.12	.740	52.97	8.00	.000	.01	.00	.01	.01
	Not assumed			52.97	7.7	.000	.01	.00	.01	.01
CH_2	Assumed	.44	.527	-54.06	8.00	.000	.00	.00	.00	.00
	Not assumed			-54.06	7.93	.000	.00	.00	.00	.00
CH_3	Assumed	.18	.679	-43.80	8.00	.000	.00	.00	.00	.00
	Not assumed			-43.80	7.99	.000	.00	.00	.00	.00

G.4.13 MTS Roy & Co. and MTO Izel

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	.92	.00	.00
	MTO Izel	5	.92	.00	.00
CH_2	MTS Roy & Co.	5	.04	.00	.00
	MTO Izel	5	.04	.00	.00
CH_3	MTS Roy & Co.	5	.05	.00	.00
	MTO Izel	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.16	.696	-2.87	8.00	.021	.00	.00	.00	.00
	Not assumed			-2.87	7.57	.022	.00	.00	.00	.00
CH_2	Assumed	.14	.721	2.26	8.00	.054	.00	.00	.00	.00
	Not assumed			2.26	7.35	.056	.00	.00	.00	.00
CH_3	Assumed	.08	.785	3.26	8.00	.012	.00	.00	.00	.00
	Not assumed			3.26	7.83	.012	.00	.00	.00	.00

G.4.14 MTS Roy & Co. and MTO Fusion Homoeopathics

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	.92	.00	.00
	MTO Fusion Homoeopathics	5	.92	.00	.00
CH_2	MTS Roy & Co.	5	.04	.00	.00
	MTO Fusion Homoeopathics	5	.03	.00	.00
CH_3	MTS Roy & Co.	5	.05	.00	.00
	MTO Fusion Homoeopathics	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	1.21	.304	-43.05	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-43.05	7.5	.000	-.01	.00	-.01	-.01
CH_2	Assumed	2.00	.195	39.76	8.00	.000	.00	.00	.00	.00
	Not assumed			39.76	6.92	.000	.00	.00	.00	.00
CH_3	Assumed	.44	.525	4337	8.00	.000	.00	.00	.00	.00
	Not assumed			43.37	7.85	.000	.00	.00	.00	.00

G.4.15 MTS Roy & Co. and MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Roy & Co.	5	.92	.00	.00
	MTS Homoeopathix	5	.92	.00	.00
CH_2	MTS Roy & Co.	5	.04	.00	.00
	MTS Homoeopathix	5	.03	.00	.00
CH_3	MTS Roy & Co.	5	.05	.00	.00
	MTS Homoeopathix	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	7.55	.025	-27.74	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-27.74	5.79	.000	-.01	.00	-.01	-.01
CH_2	Assumed	1.58	.245	27.88	8.00	.000	.00	.00	.00	.00
	Not assumed			27.88	5.43	.000	.00	.00	.00	.00
CH_3	Assumed	32.94	.000	12.72	8.00	.000	.00	.00	.00	.00
	Not assumed			12.72	4.44	.000	.00	.00	.00	.00

G.4.16 MTS Helios and MTO Izel

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	.91	.00	.00
	MTO Izel	5	.92	.00	.00
CH_2	MTS Helios	5	.04	.00	.00
	MTO Izel	5	.04	.00	.00
CH_3	MTS Helios	5	.05	.00	.00
	MTO Izel	5	.05	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.51	.494	-48.36	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-48.36	6.89	.000	-.01	.00	-.01	-.01
CH_2	Assumed	.63	.451	47.03	8.00	.000	.00	.00	.00	.00
	Not assumed			47.03	6.99	.000	.00	.00	.00	.00
CH_3	Assumed	.01	.942	43.75	8.00	.000	.00	.00	.00	.00
	Not assumed			43.75	7.89	.000	.00	.00	.00	.00

G.4.17 MTS Helios and MTO Fusion Homoeopathics

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	.91	.00	.00
	MTO Fusion Homoeopathics	5	.92	.00	.00
CH_2	MTS Helios	5	.4	.00	.00
	MTO Fusion Homoeopathics	5	.03	.00	.00
CH_3	MTS Helios	5	.05	.00	.00
	MTO Fusion Homoeopathics	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	2.63	.144	-90.51	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-90.51	6.82	.000	-.01	.00	-.01	-.01
CH_2	Assumed	3.05	.119	82.10	8.00	.000	.01	.00	.01	.01
	Not assumed			82.10	6.54	.000	.01	.00	.01	.01
CH_3	Assumed	.09	.770	83.52	8.00	.000	.01	.00	.01	.01
	Not assumed			83.52	7.91	.000	.01	.00	.01	.01

G.4.18 MTS Helios and MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTS Helios	5	.91	.00	.00
	MTS Homoeopathix	5	.92	.00	.00
CH_2	MTS Helios	5	.04	.00	.00
	MTS Homoeopathix	5	.03	.00	.00
CH_3	MTS Helios	5	.05	.00	.00
	MTS Homoeopathix	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	11.10	.010	-59.63	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-59.63	5.27	.000	-.01	.00	-.01	-.01
CH_2	Assumed	2.25	.172	57.48	8.00	.000	.01	.00	.01	.01
	Not assumed			57.48	5.19	.000	.01	.00	.01	.01
CH_3	Assumed	35.23	.000	27.00	8.00	.000	.01	.00	.01	.01
	Not assumed			27.00	4.46	.000	.01	.00	.01	.01

G.4.19 MTO Izel and MTO Fusion Homoeopathics

Group Statistics

Method		*N55*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Izel	5	.92	.00	.00
	MTO Fusion Homoeopathics	5	.92	.00	.00
CH_2	MTO Izel	5	.04	.00	.00
	MTO Fusion Homoeopathics	5	.03	.00	.00
CH_3	MTO Izel	5	.05	.00	.00
	MTO Fusion Homoeopathics	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	.22	.651	-36.23	8.00	.000	-.01	.00	-.01	-.01
	Not assumed			-36.23	8.00	.000	-.01	.00	-.01	-.01
CH_2	Assumed	.41	.539	33.61	8.00	.000	.00	.00	.00	.00
	Not assumed			33.61	7.90	.000	.00	.00	.00	.00
CH_3	Assumed	.09	.769	37.40	8.00	.000	.00	.00	.00	.00
	Not assumed			37.40	8.00	.000	.00	.00	.00	.00

G.4.20 MTO Izel and MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Izel	5	.92	.00	.00
	MTS Homoeopathix	5	.92	.00	.00
CH_2	MTO Izel	5	.04	.00	.00
	MTS Homoeopathix	5	.03	.00	.00
CH_3	MTO Izel	5	.05	.00	.00
	MTS Homoeopathix	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	3.82	.087	-24.31	8.00	.000	-.01	.00	-.01	.00
	Not assumed			-24.31	6.67	.000	-.01	.00	-.01	.00
CH_2	Assumed	.86	.381	24.79	8.00	.000	.00	.00	.00	.00
	Not assumed			24.79	6.44	.000	.00	.00	.00	.00
CH_3	Assumed	28.00	.001	11.48	8.00	.000	.00	.00	.00	.00
	Not assumed			11.48	4.58	.000	.00	.00	.00	.00

G.4.21 MTO Fusion Homoeopathics and MTS Homoeopathix

Group Statistics

Method		*N*	*Mean*	*Std. Deviation*	*S.E. Mean*
H_2O	MTO Fusion Homoeopathics	5	.92	.00	.00
	MTS Homoeopathix	5	.92	.00	.00
CH_2	MTO Fusion Homoeopathics	5	.03	.00	.00
	MTS Homoeopathix	5	.03	.00	.00
CH_3	MTO Fusion Homoeopathics	5	.04	.00	.00
	MTS Homoeopathix	5	.04	.00	.00

Independent Samples Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
	Equal Variances	*F*	*Sig.*	*t*	*df*	*Sig. (2-tailed)*	*Mean Diff.*	*Std. Error Difference*	95% Confidence Interval of the Difference	
									Lower	*Upper*
H_2O	Assumed	4.00	.081	2.87	8.00	.021	.00	.00	.00	.00
	Not assumed			2.87	6.74	.025	.00	.00	.00	.00
CH_2	Assumed	.33	.581	-.59	8.00	.571	.00	.00	.00	.00
	Not assumed			-.59	6.88	.574	.00	.00	.00	.00
CH_3	Assumed	31.72	.000	-2.29	8.00	.051	.00	.00	.00	.00
	Not assumed			-2.29	4.57	.076	.00	.00	.00	.00