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A Bayesian Hierarchical ^{13}COBT to Correct Estimates Associated with a Delayed Gastric Emptying

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Abstract**—**The use of a Bayesian Hierarchical Model (BHM) to interpret breath measurements obtained during a ^{13}C Octanoic Breath Test (^{13}COBT) is demonstrated. The statistical analysis was implemented using WinBUGS, a commercially available computer package for Bayesian inference. A hierarchical setting was adopted where poorly defined parameters associated with a delayed Gastric Emptying (GE) were able to "borrow" strength from global distributions. This is proved to be a sufficient tool to correct model's failures and data inconsistencies apparent in conventional analyses employing a Non-linear least squares technique (NLS). Direct comparison of two parameters describing gastric emptying (t_{lag} -lag phase, $t_{1/2}$ -half emptying time) revealed a strong correlation between the two methods. Despite our large dataset ($n = 164$), Bayesian modeling was fast and provided a successful fitting for all subjects. On the contrary, NLS failed to return acceptable estimates in cases where GE was delayed.

Keywords**—**Bayesian hierarchical analysis, ^{13}COBT, Gastric emptying, WinBUGS.

I. INTRODUCTION

GAMMA Scintigraphy is widely recognised as the 'gold standard' method for measuring Gastric Emptying (GE) in humans. The clinical procedure and different variations of the protocol have been well described [1][2], but essentially the test requires the subject to ingest a radio-labelled meal and in the following few hours a number of images of the quantity of radiolabel retained in the stomach are obtained. These measurements are used to identify a mathematical model (power exponential function) [3] from which the time of the maximum emptying rate ($t_{lag(s)}$) and the time at which half of the ingested label substrate has left the stomach ($t_{1/2(s)}$) can be derived.

However, use of the test raises several ethical issues such as exposure of the patient to a significant amount of radiation, and its application in paediatrics or pregnancy. Furthermore there is a number of inconsistencies between reported studies (meal composition, meal form, display methodology, quantitative parameters) that have led to a need for the development of an alternative test capable of overcoming these issues.

The ^{13}C-octanoate acid breath test (^{13}COBT) was first introduced by Ghoos [4] as an indirect, low cost and ethically acceptable method to measure gastric emptying in man. Due to its nature the test is suitable for repetitive studies and in all populations. The octanoate (in common with all medium chain fatty acids) passes unchanged through the stomach and enters the small intestine where is it rapidly absorbed and then transported to the liver where is immediately oxidised into CO_2 and water. Subsequently, the only limiting factor for its metabolism is the time interval that it actually resides in the stomach.

The standard protocol for the ^{13}COBT requires the ingestion of a labelled solid meal followed by a number of breath measurements of the instantaneous recovery of the labelled $^{13}CO_2$. By fitting a mathematical model to a set of breath measurements two key parameters can be obtained, known as the half and lag time ($t_{1/2(b)}$ and $t_{lag(b)}$ respectively).

Although the ^{13}COBT has been exploited in many studies [5][6], the test has not yet achieved universal acceptance, due mainly to its inability to provide parameters which are directly comparable to those from gamma scintigraphy. The reason for this is that firstly approximately half of the recovery of the label is lost in the tricarboxylic acid cycle [7]-[10], and secondly its appearance in breath is delayed by its passage through the bicarbonate pools, which leads to a reduced and blunted signal in comparison with scintigraphy (Fig. 1).

Recently, a "self-correction" mathematical model has been proposed to estimate GE using breath measurement as the input data [11]. Model parameters were found to highly correlate with the ones from the gamma scintigraphy test although its identification requires that the $^{13}CO_2$ in breath is well defined and that GE is not severely delayed (Fig. 2) as frequently happens in many pathological states such as obesity and type 2 diabetes mellitus [12][13].

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The application of some prior knowledge to the statistical inference of the parameters, within the context of a Bayesian framework, is suggested as a sufficient tool to overcome the deficiency in the data when GE is delayed. Bayesian techniques allow the investigator to include some information

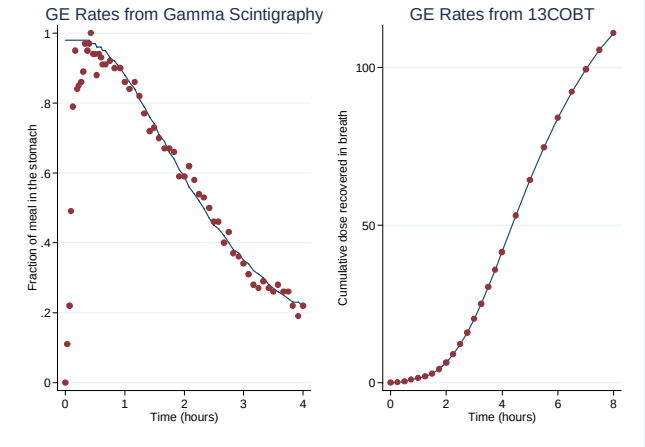

Fig. 1 Gastric emptying patterns as illustrated by a Gamma Scintigraphy Test(left panel) and a ^{13}COBT (right panel).

before the analysis is commenced. In practice this is done by defining probability distributions for the model's macroparameters which are subsequently updated by the measurements (data), producing a full probability model. This can be done either by allowing these parameters to vary freely individually or by choosing to allow them to share a number of common properties and characteristics in the context of a hierarchical setting.

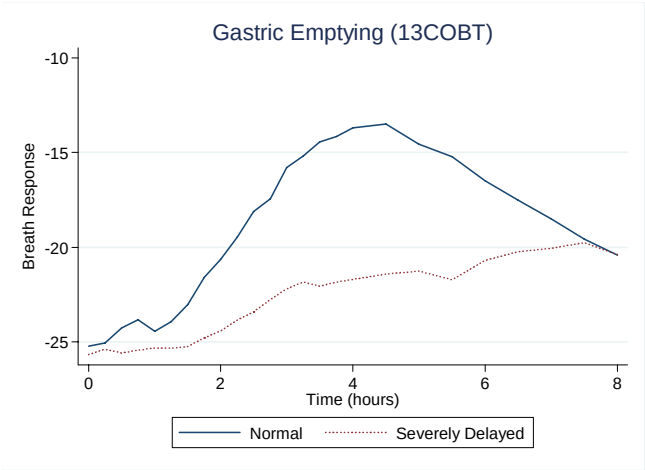

Fig. 2 Illustration of a normal and a severely delayed Gastric Emptying.

This work aims to introduce a Bayesian Hierarchical Model (BHM) into the ^{13}COBT for the estimation of GE in a wide range of cases comprising well- and poorly- defined breath recovery curves. For this purpose we exploited a large dataset from several studies conducted in our laboratories and which have already been reported in the literature.

II. METHODS

A. Dataset

A number of studies previously conducted at our laboratories have been reanalysed for this work (Table I).

TABLE I
STUDIES USED IN THE STATISTICAL ANALYSIS

No	Study	Breath Datasets	Ref.
1	Frequent Feasibility Feeding study	16	[14]
2	Frequent Feeding study	32	[14]
3	Obesity Study	32	[15]
4	Meal Size study	36	[16]
5	Meal Composition study	48	[16]

In all a total of 164 sets of breath test data obtained from a large population (lean and obese subjects) were reanalyzed simultaneously using a Bayesian hierarchical model. The reader is referred to the citations in Table I for more information regarding the protocols.

B. Generic Protocol for the Breath Test

The studies were performed in the volunteer suite at MRC Human Nutrition Research (MRC-HNR), Cambridge, UK and were approved by the Local Research Ethics Committee. All the reported investigations were performed on healthy adults with no history of gastrointestinal disorder. Informed written consent to the tests was obtained from each subject following the receipt of written explanations of the protocol and purpose of the study.

C. Study-day protocol

Subjects were asked to abstain from alcohol and strenuous physical activity, and to fast from 8.00 pm on the day before being studied. To prevent dehydration a single glass of water was allowed on waking on the morning of the study. On arrival at the MRC-HNR height (without shoes) was measured to the nearest 1 cm and weight determined to the nearest 0.01 kg for the estimation of basal metabolic rate (BMR) [17]. The ^{13}COBT using an egg-based meal, with a standard energy content of 2MJ. The egg was separated and a standard dose of 100μL of ^{13}C-octanoic acid (621μmole) added to the yolk. The egg was then dry-fried in a non-stick pan, and served with 3 slices of toasted bread spread with a total of 10g butter, 100mL of orange juice and 100mL water. Subjects were asked to consume the meal in less than ten minutes.

Basal samples of breath were collected prior to the meal being consumed and continued for six hours. For the first four hours samples were taken every fifteen minutes, and every half hour for the final two-hour period. All breath samples were collected in duplicate. Isotope analysis was performed using isotope ratio mass spectrometry (IRMS), with a reference gas traceable to an international standard V-(PDB).

The calculations were performed as follows:

CO_2 production rate (mole/hr),

$$F_{CO_2}(n) = 0.04518\, W(n)^{0.5378} H(n)^{0.3964} \quad (1)$$

from the formula given by [18], where $W(n)$ is the weight (kg) and $H(n)$ the height (m) for the n^{th} subject.

The isotope data was fitted to curves according to (2)

$$\delta(n,t) = \delta_b(n) + \frac{1000 \cdot d}{(PDB) \cdot F_{CO_2}(n)} F_\infty(n) k(n) \beta(n) \left(1 - \exp\{-k(n)t\}\right)^{\beta(n)-1} \cdot \exp\{-k(n)t\} \quad (2)$$

where $F_\infty(n)$ (the fraction of the dose recoverable in breath), $k(n)$, and $\beta(n)$ are parameters of the fit for the n^{th} subject, d is the isotope dose given, PDB the isotopic composition of the international standard, and $\delta(n,t)$ and $\delta_b(n)$ (‰) the measured isotopic compositions with respect to PDB for each of the breath test at time t and in the basal state respectively.

From the coefficients of the fit two parameters descriptive of the gastric emptying process were calculated [19]

$$t_{1/2}(n) = \frac{-Ln\left(1 - 2^{-1/\beta(n)}\right)}{k(n)} \quad (3)$$

$$t_{lag}(n) = \frac{Ln\{\beta(n)\}}{k(n)} \quad (4)$$

D. Bayesian Hierarchical Analysis

Bayesian modelling was performed using a conditional BHM consisting of the available breath data ($k = 1,2....164$) and a degree of prior information embedded to the unknown parameter vector $\theta(n) = \{F_\infty(n), k(n)\}$ from which the two parameters of interest $\left\{t_{1/2}^{BAY}(n), t_{lag}^{BAY}(n)\right\}$ were then estimated. According to Bayesian inference, prior information was updated form the available dataset to formulate conditional probability distributions known as *posteriors*. Posterior distributions are weighted products between the likelihood of the data and the prior information according to Bayes Theorem [20]

$$p(\theta(n) \mid y(n)) = \frac{p(y(n) \mid \theta(n)) p(\theta(n))}{p(y(n))} \quad (5)$$

where $y(n)$ is the available data (breath test measurement $\delta(n,t)$ for the n^{th} subject obtained at time $t = 1,2.....25$), θ is the unknown parameter vector, $p(\theta(n))$ is the probability vector consisting of the prior beliefs, $p(y(n) \mid \theta(n))$ is the likelihood, $p(y(n))$ is a scaling factor, and $p(\theta(n) \mid y(n))$ is the posterior probability distribution vector. By omitting the scaling factor $p(y(n))$ which is independent on θ one can obtain a reduced form of Bayes theorem,

$$p(\theta(n) \mid y(n)) \propto p(y(n) \mid \theta(n)) p(\theta(n)) \quad (6)$$

We let individual parameters of fit $F_\infty^{BAY}(n), k^{BAY}(n)$ depending on other hyperparameters $F_\infty^{BAY}(p), k^{BAY}(p)$ which were assigned vague non-informative priors (p for population). The richness of our data and the fact that all subjects underwent the same protocol, allowed us to build a hierarchical setting with parameters borrowing strength from each other, as illustrated from the graphical model in Figure 3.

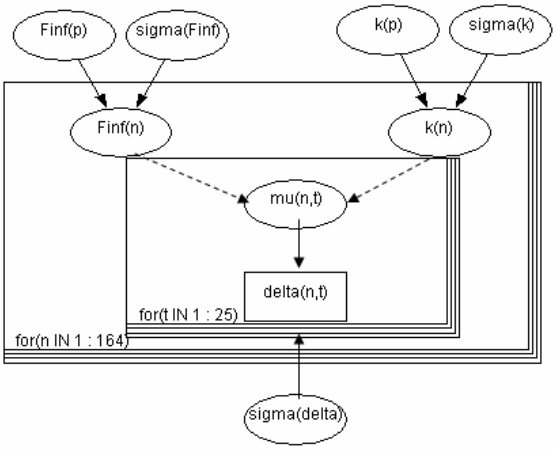

Fig. 3 Graphical (Doodle) representation of the Bayesian Hierarchical Model used in this study

The commercially available computer package WIBUGS [21] was used to implement the model. WinBUGS was used to generate two Markov Chains [22] comprising 6000 samples for each of the unknown parameters. The first 1000 samples were used as the "burn-in" and the rest (5000) for the posterior sampling. The entire process took approximately 34 minutes in a standard desktop workstation.

E. BHM versus NLS

Comparison of results from individual studies with their Non-linear least squares (NLS) counterparts previously obtained using MS Excel was used to assess the performance

of the Bayesian method. Parameter estimates are reported as posterior means ± standard deviation (SD) for the BHM $\left(F_{\infty}^{BAY}(p), k^{BAY}(p), t_{1/2}^{BAY}(n), t_{lag}^{BAY}(n)\right)$ and point estimates ± standard deviations for the NLS $\left(t_{1/2}^{NLS}(n), t_{lag}^{NLS}(n)\right)$. Pearson correlation was used to assess the agreement of the results obtained by the two methods and statistical significance using a 5% significance level was determined using a paired t-test.

III. RESULTS

The global distribution defining the parameters $F_{\infty}^{BAY}(p)$, and $k^{BAY}(p)$ were all found to be near-normally distributed (mean and median coincident), with symmetrical confidence limits. $F_{\infty}^{BAY}(p)$ was found to be 42.9±0.8% (mean ± SD) and $k^{BAY}(p)$ =0.511±0.012hr^{-1}. The ranges of the individual gastric emptying time parameters were 1.66hr< t_{lag}^{BAY} <7.25hr, and 2.23hr< $t_{1/2}^{BAY}$ <8.07hr. In general the standard deviation of the individual value of the parameter was found to increase with its absolute value, with the average coefficient of variation (c.v.) being 2.2%, and 2.7% for t_{lag}^{BAY} and $t_{1/2}^{BAY}$ respectively. The β parameter which is also obtained from the fitted curve and is closely associated to the derivation of the key parameters (t_{lag} , $t_{1/2}$) was successfully obtained by both the BHM and the NLS methods. It is worth mentioning that β derived from the Bayesian approach (β^{BAY}) was much larger for obese subjects compared to its values associated with leans.

Bayesian analysis and the BHM implemented in WinBUGS returned a successful fit and derived estimates for all subjects in contrast to the NLS which returned unsatisfactory fitting for all individuals with a delayed GE. A representative illustration of the Bayesian model fit is shown in Fig. 4.

Comparison of the two estimates (t_{lag}), and ($t_{1/2}$) from the NLS and Bayesian methods is indicative of the 'borrowing of strength' effect featured in the hierarchical analysis (Fig. 5). Both parameters obtained from the two methods were high correlated with regression lines being identical to the unity (slopes equal to 1, $r = 0.84$ and $r = 0.82$ for t_{lag} and $t_{1/2}$ respectively). Further paired t-tests did not reveal any statistical difference for t_{lag} and $t_{1/2}$ derived from the two methods.

IV. DISCUSSION

In this paper it is clearly demonstrated that Bayesian analysis is a superior approach to estimate gastric emptying in a large dataset of subjects obtained from different studies in the literature. Furthermore, it was observed that the Bayesian hierarchical model is capable to fully interpret breath data obtained from an ^{13}COBT even in cases where GE was severely delayed. Moreover, the Bayesian method was found adequate to fully overcome problematic issues associated to the conventional NLS such as failures in model's fitting, inconsistent data arising when GE is delayed and large uncertainty across the parameters.

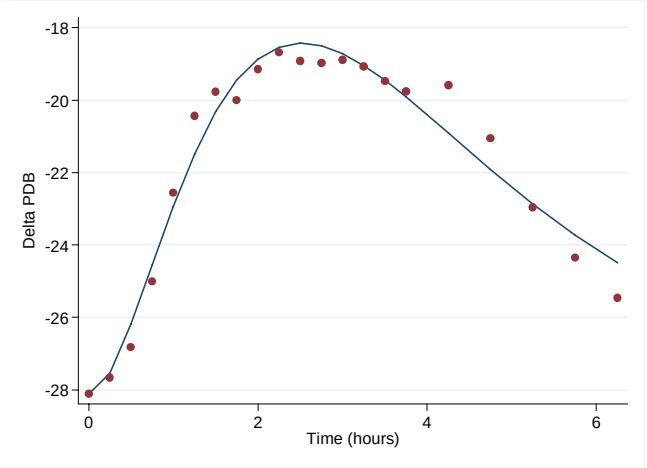

Fig. 4 Raw breath test results and the Bayesian fit.

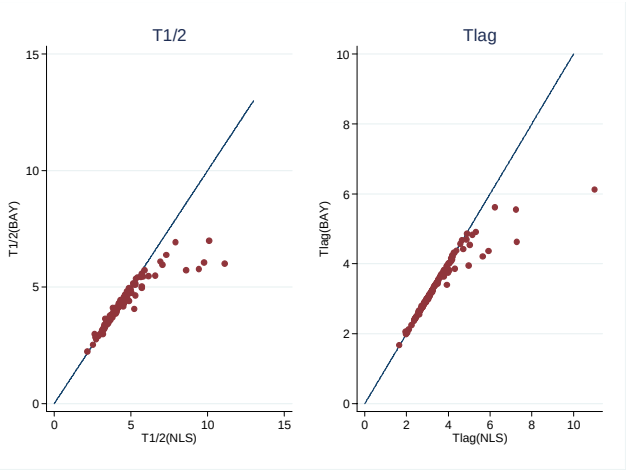

Fig. 5 Regression lines for $t_{1/2}$ (left panel) and t_{lag} (right panel). A strong agreement was observed between the two methods.

The use of the commercially available computer package WinBUGS allowed the satisfactory generation of two Markov Chains from which we derived posterior means with the associated shape of uncertainty (probability distributions) for each of the parameters. Furthermore, the program allowed the automatic generation of the limits of credibility for all parameters.

The adoption of a hierarchical setting was shown to reduce the labour of the computational analysis compared to the classical individual approach. The intensiveness and complexity of Bayesian analysis has been mentioned many times in the literature [23]. In the present analysis, the time required for the model's fit was slightly more than 30 minutes which compares favourably to other methods particularly when bearing in mind all the extra features associated to the WinBUGS package.

In addition, there was strong correlation between the two methods especially in cases where the gastric emptying was not ill-posed. Similar regression figures have been also reported by others [4]. However, the significant advancement of the Bayesian method was clearer when dealing with subjects with a severely delayed GE. In contrast to the NLS, Bayesian method returned estimates and a successful modelling for all subjects. Bayesian analysis was able to discriminate between lean and obese subjects based on their β parameter. It was found that β^{BAY} parameter was greater in obese people who feature a delayed GE, something that is in concordance with other studies in the literature [3].

The main advantage of the Bayesian approach is the ability to provide theoretically self-consistent and plausible estimates of individual and population parameters, albeit with extra care being required during data preparation.

In summary Bayesian hierarchical methods provide a robust and reliable way to analyses the gastric emptying breath test. The methods avoid parameter estimation failure in cases where data quality is sub-optimal, yet reproduce the results of standard nom-linear least squares methods when the data is well-behaved. We would recommend that the WinBUGS package be adopted routinely for the estimation of gastric emptying by ^{13}C-octanoate breath test.

REFERENCES

[1] Mariani G, Boni G, Barreca M, "Radionuclide gastroesophageal motor studies", *J Nucl Med*, vol.45(6), pp:1004-1028, 2004.

[2] Abell TL, Camilleri M, Donohoe K, "American Neurogastroenterology and Motility Society and the Society of Nuclear Medicine. Consensus recommendations for gastric emptying scintigraphy: a joint report of the American Neurogastroenterology and Motility Society and the Society of Nuclear Medicine", *J Nucl Med Technol*, vol.36(10), pp:44-54, 2008.

[3] Siegel JA, Urbain JL, Adler LP, Charkes ND, Maurer AH, Krevsky B, Knight LC, Fisher RS, Malmud LS, "Biphasic nature of gastric emptying", *Gut*, vol.29, pp:85-89, 1988.

[4] Ghoos YF, Maes BD, Geypens BJ, Mys G, Hiele MI, Rutgeerts PJ, Vantrappen G, "Measurement of Gastric Emptying Rate of Solids my Means of a Carbon-Labeled Octanoic Acid Breath Test", *Gastroenterology*, vol.104, pp:1640-1647,1993.

[5] Maes BD, Ghoos YF, Geypens BJ, Mys G, Hiele MO, Rutgeerts PJ, Vantrappen G, "Combined Carbon-13-Glycine/Carbon-14- Octanoic Acid Breath Test to Monitor Gastric Emptying Rates of Liquids and Solids", *J Nucl Med*, vol.35, pp:824-831, 1994.

[6] Maes BD, Mys G, Geypens BJ, Evenepoel P, Ghoos YF, Rutgeerts PJ, "Gastric emptying flow curves separated from carbon-labeled octanoic acid breath test results", *Gastrointest Liver Physiol*, vol.38, pp:169-175, 1998.

[7] Sidossis LS, Coggan AR, Gastaldelli A, "A new correction factor for use in tracer estimations of plasma fatty-acids in non-anesthetized pigs", *Am J Physiol - Endocrinol Metab*, vol.32, pp:649-656, 1995.

[8] Schrauwen P, van Aggel-Leijssen DPC, Lichtenbelt WDV, "Validation of the [1,2-C-13]acetate recovery factor for correction of [U-C-13]palmitate oxidation rates in humans", *J Physiol Lond*, vol.513, pp:215-223, 1998.

[9] van Hall G, "Correction factors for C-13-labelled substrate oxidation at whole-body and muscle level", *Proc Nutr Soc*, vol.58, pp:979-986, 1999.

[10] Schrauwen P, Blaak EE, van Aggel-Leijssen DPC, "Determinants of the acetate recovery factor: implications for estimation of [13-C] substrate oxidation", *Clin Sci*, vol.98, pp:587-592, 2000.

[11] Bluck LJ, Coward WA, "Measurement of gastric emptying by the ^{13}C-octanoate breath test-rationalization with scintigraphy", *Physiol Meas*, vol.27, pp:279-289, 2006.

[12] Samsom M, Bharucha A, Gerich JE, Herrmann K, Limmer J, Linke R, Maggs D, Schirra J, Vella A, Worle HJ, Goke B, "Diabetes mellitus and gastric emptying: questions and issues in clinical practice", *Diabetes Metab Res Rev*, vol.25, pp:502-514, 2009.

[13] Horowitz M, Collins PJ, Cook DJ, Harding PE, Shearman DJC, "Abnormalities of Gastric Emptying in Obese Patients", *Int J Obes*, vol.7, pp:415-421, 1983.

[14] Jackson SJ, Leahy FE, Jebb SA, Prentice AM, Coward WA, Bluck LJC, "Frequent feeding delays the gastric emptying of a subsequent meal", *Appetite*, vol.48(2), pp:199-205, 2007.

[15] Jackson SJ, Leahy FE, McGowan AA, Bluck LJC, Coward WA, Jebb SA, "Delayed gastric emptying in the obese: an assessment using the non-invasive C-13-octanoic acid breath test", *Diabetes Obes Metab*, vol.6(4), pp:264-270, 2004.

[16] Jackson SJ, Bluck LJC, Coward WA, "Use of isotopically labelled octanoic acid to assess the effect of meal size on gastric emptying", *Rapid Commun Mass Spectrom*, vol. 18(10), pp:1003-1007, 2004.

[17] Schofield W, "Predicting Basal Metabolic Rate, New Standards and Review of Previous Work", *Hum Nutr Clin Nutr*, vol.39C, pp:5-41, 1985.

[18] Evenepoel P, Geypens B, Luypaerts A, Hiele M, Ghoos Y, "Digestibility of Cooked and Raw Egg Protein in Humans as Assessed by Stable Isotope Techniques", *Journal of Nutrition*, vol.128, pp:1716-1722, 1998.

[19] Schommartz B, Ziegler D, Schadewaldt P, "Significance of diagnostic parameters in [C-13]octanoic acid gastric emptying breath tests", *Isotopes Environ Health Stud*, vol.34, pp:135-143, 1998.

[20] Gelman A, Carlin JB, Stern HS, Rubin DB, "Bayesian Data Analysis", *Chapman & Hall*, TX: Statistical Science, 2004.

[21] Lunn DJ, Thomas A, Best N, Spiegelhalter D, "WinBUGS - a Bayesian modelling framework: concepts, structure and extensibility", *Stat Comput*, vol.10, pp:325-337, 2000.

[22] Gilks WR, Richardson S, Spiegelhalter DJ, "Markov Chain Monte Carlo in Practice", *Chapman & Hall*, TX: Interdisciplinary Statistics, 1996.

[23] Pilloneto G, Sparacino G, Magni P, Bellazzi R, Cobelli C, "Minimal model Si=0 problem in NIDDM subjects: nonzero Bayesian estimates with credible confidence intervals", *Am J Physiol Endrocrinol Metab*, vol.282, pp:564-573, 2000.