Original article

Conception and development of a bibliographic database of blood nutrient fluxes across organs and tissues in ruminants: data gathering and management prior to meta-analysis

Jean VERNET*, Isabelle ORTIGUES-MARTY

Unité de Recherches sur les Herbivores, INRA, Theix, 63122 Saint-Genès-Champanelle, France

Abstract – In the organism, nutrient exchanges among tissues and organs are subject to numerous sources of physiological or nutritional variation, and the contribution of individual factors needs to be quantified before establishing general response laws. To achieve this, meta-analysis of data from publications is a useful tool. The objective of this work was to develop a bibliographic database of nutrient fluxes across organs and tissues of ruminant animals (Flora) under Access using the Merise method. The most important criteria for Flora were the ease to relate the various information, the exhaustivity and the accuracy of the data input, a complete description of the diets, taking into account the methods of the methodological procedures of measurement and analysis of blood nutrients and the traceability of the information. The conceptual data model was built in 6 parts. The first part describes the authors and source of publication, and the person in charge of data input. It clearly separates and identifies the experiments, the groups of animals and the treatments within a publication. The second part is concerned with feeds, diets and their chemical composition and nutritional value. The third and fourth parts describe the infusion of any substrates and the methods employed, respectively. The fifth part is devoted to the results of blood flows and nutrient fluxes. The sixth part gathers miscellaneous experimental information. All these parts are inter-connected. To model this database, the Merise method was utilised and 26 entities and 32 relationships were created. At the physical level, 93 tables were created, corresponding, for the majority, to entities and relationships of the data model. They were divided into reference tables (n = 65) and data tables (n = 28). Data processing was developed in Flora and included the control of the data, generic calculations of unknown data from given data, the automation of the estimation of the missing data or the chemical composition of the diets. It also included the construction of tables for meta-analyses and the study of the variations of several factors within publications (pre-coding of meta-analyses). Overall, the system was built to facilitate the gathering, input, validation, management and retrieval of data from publications.

bibliography / database / Merise / Access / net nutrient fluxes

1. INTRODUCTION

In animal nutrition, especially in the areas of tissue or cellular metabolism, new questions are arising. New information needs to be integrated and related to previous information in order to integrate knowledge from different tissues and organs. Such integration is required to account for nutrient metabolism and their

---

* Corresponding author:
 jean.vernet@clermont.inra.fr

regulatory factors [1, 2]. The present work belongs to a general research program which is aimed at building a generalised and quantitative representation of nutrient exchanges between and within tissues and organs in the animal.

A pre-requisite to the construction of models [3] is the knowledge of response laws [4]. Response laws can be established using a meta-analysis approach after having gathered information of diverse origin [5, 6].

The general principles and advantages of databases have already been discussed by [7]. In particular, computerised relational databases are powerful tools which can relate (by way of common identifiers) and process a wide range of information.

A large corpus of published data on net fluxes of nutrients originates from experiments conducted using multicatheterised animals. The animal species were generally ruminants, in particular the ovine and bovine. The organs and tissues of focus were the portal drained viscera, the liver, and the hindlimb or the muscle [8]. The nutrients under study were energetic and nitrogenous. The development of the present database, named Flora (FLuxes of nutrients through ORgans and tissues in Ruminant Animals), constituted an original and wider development of a previous experimental database [9]. Flora was specifically conceived to gather data averaged from groups of animals and from a series of measurements, and to widely cover different experimental plans and objectives (nutritional, infusion of substrates, physiological stages). Since the nature of the reported data and their presentation could vary with the publication it was decided to input data in a similar and uniform matter. Therefore, the objective of the present paper was to describe the conception and development of a computerised database (Flora) which gathered published data in the field of animal nutrition, tissue and or-

gan metabolism, for subsequent use by a statistical meta-analytical approach.

2. MATERIALS AND METHODS

2.1. Specifications for the construction of the Flora database

The first specification was to gather all the information which was available in the publication.

The second specification was to store significant quantities of information, in a reliable way, and to easily integrate and retrieve information.

The third specification was related to the validation of information. The data were to be verified with that reported in the publication and had to be quantitatively coherent within the publication (i.e. concentrations, blood flow and blood nutrient fluxes). In addition, occasionally, the results seemed aberrant or influent (blood flow and nutrient fluxes, characteristics of the diets). Therefore, we decided to set up a compulsory step of data validation before data input took place.

The fourth specification delt with the fact that the database utilised nutritional studies. Parameters of the chemical composition and the nutritional value of the feeds and diets were taken into account, since they are known to play a role in the metabolism of nutrients at the organ and tissue level [10,11]. Whenever this vital information was missing, it was decided to estimate it using a specific protocol (see Sect. 3.1.3).

The fifth specification was to take into account the potential complexity of infusion procedures. For example, the simultaneous infusion of different substrates at different anatomical sites [12].

The sixth specification dealt with the different measurement and analytical methods used by the authors in order to explain any variation observed between

nutrient fluxes from different publications. Because of the importance of the methodological approaches utilised on the results and subsequent interpretation of data [2, 13, 15], it was decided to include information on the methodology used by the authors in Flora.

The seventh specification concerned the traceability of the results. The results may not have been directly reported in the publication. Consequently, "reported" or "calculated" or "estimated" was assigned to accompany each result.

The eighth specification took into account the intra-trial variability to allow to use it as a weighting factor of the variables in meta-analyses [16–19].

The ninth specification was related to the choice of units. The units often differed between publications. Thus, the choice of an appropriate unit for each type of result was necessary for uniformity of data.

Finally, the tenth requirement concerned the ease of data input or retrieval into, or from Flora. It was decided to input the data into Excel [20], since this program is well known and easy to use. Once this was done, the data were then transferred into Access [21].

2.2. Specifications for data management

Initially, there were several criteria required for data management. Data were verified, checked for missing values after the loading step, and the coherence between the various types of information was verified.

Secondly, in order to have an exhaustive database, it was necessary to make calculations from existing data in the publication (e.g. to calculate concentrations from net blood fluxes). These calculations were made after the input and loading steps.

Finally, it was also important to check and describe the data present in Flora and the metadispositive of each explicative X variable. The latter makes it possible to know the ranges of publications, and inside these ranges the zones with large numbers of observations and those with missing data [22].

In order to answer specific research questions (e.g. "the evolution of energy fluxes of nutrients in the portal vein according to the feeding conditions in sheep") it was necessary to construct new tables for each meta-analysis.

The coding of data was a significant step. It consisted in defining and applying a code [code on X, e.g. dry matter intake (DMI)] based on only one source of variation (i.e. a variation on X and no variations on other factors). The coding concerned successively the following: 1/ publications and 2/ within publication groups of treatments (sets of treatments with a homogeneity of variation between them). The initial coding (1/) was done automatically in Flora. When the number of publications was significant, $n > 100$, coding was largely facilitated by an automation (pre-coding step). After the initial coding was completed, publications were then coded manually after thorough examination, by subdividing them into treatment groups (2/).

2.3. The publications used in Flora

The publications included in Flora generally only detailed one experiment. However, some publications are composed of several separate experiments [23].

The majority of the experimental animals described in the sample of publications were bovine (70%), with ovine making up the balance (30%). The description of the animals in the model were based on groups of animals, rather than individual animals. The group of animals corresponded to the set of animals of a given physiological state which was used in a specific experiment and for which the results of fluxes were reported. In some cases

the same groups of animals were found in several publications, when the authors reported different results from the same experiment e.g. [24, 25]. The animals used in a given experiment were subdivided into several groups on the basis of their physiological status (e.g. growth, lactation, ...). All animals were surgically equipped with catheters in an artery and in several veins, e.g. the portal and hepatic veins.

The publications were described according to 4 categories of treatments: 1/ dietary treatments, 2/ those utilising infused substrates, 3/ physiological states, and 4/ miscellaneous treatments. With the first type of treatment, the diets differed by their ingredient and/or chemical composition or by the modes of feed preparation. With the second type of treatment, substrates were either infused by way of intra-digestive fistula and/or intra-venous catheters to provide specific energetic or nitrogenous supplements, (e.g. volatile fatty-acids or casein). With the third type of treatment, a physiological state was described according to different stages, e.g. growing animals were studied at different age and body weight [26]. Finally, in a small number of cases, other treatments were studied, e.g. to compare resting and exercised animals exposed to cold and thermoneutral environments [27].

The experimental designs which were most frequently reported were factorial, latin squares or split-plot designs. In the case of factorial designs, all the main factors and possible interactions between them were studied.

Measurements of blood flow and blood sampling often took place during the last day of treatment. Blood flows were either measured by flow probes [28] or by dilution methods [14, 29]. Additionally, blood samples were collected from the catheter in most cases by spot sampling rather than continuous sampling, for immediate or further blood metabolite determination. Rumen fluid could also be collected for im-

mediate or further determination of rumen fluid parameters.

Metabolite and chemical determinations were carried out in feed, blood, plasma or rumen fluid samples. They were based mainly on enzymatic, spectrophotometric and chromatographic methods.

Data on net fluxes of blood or plasma nutrients included concentrations, arteriovenous concentration differences, net nutrient fluxes and fractional extraction of either energetic nutrients (e.g. glucose), and/or nitrogenous nutrients (e.g. ammonia-nitrogen). The results concerned with rumen fluid parameters included pH, ammonia and individual volatile fatty-acid concentrations. Statistical error terms were generally given for all treatments utilising the terms "standard error" or "standard error of the mean".

2.4. Data modelling

It was very important to accommodate all data to be recorded and all data processing [30] when constructing the database structure, represented by the conceptual data model [31]. The Merise modelling method was chosen, since it is well known and it has been used successfully in agricultural research [7, 32].

3. RESULTS

3.1. Preliminary steps for the construction of the conceptual database model and the Flora database

3.1.1. Identification and description of the data to be input

The first requirement set for Flora was aimed at identifying all the necessary information that had to be entered into the database. This information was identified

from 2 single publications that differed in their experimental design and provided a detailed description of their Materials and Methods [12, 24]. The mandatory information included the authors and their research teams, the full reference, the description of the animals within treatment groups, the treatments, the feeds and diets, the infused substrates, the time-course of the experiment, the sampling protocol, the statistical analyses, analytical methods and the results (blood flows, blood nutrients, rumen parameters and zootechnical performances).

Once the list of the mandatory information was compiled, it was considered important to be able to easily and rapidly sort the 4 main categories of experimental treatments, namely "dietary", "substrate infusion", "physiological stage" and "miscellaneous" (this latter included cases like "sheared vs. covered" or "resting vs. exercised". Therefore, it was decided to add an additional property "type of treatment" to the model.

For the infused substrates category, the description of the infusion was relatively complex. For example, several substrates could be infused at the same time, or there could be different sites of infusion. Therefore, the infused substrate mixture was described according to a hierarchical logic in order to be able to contend with one or several single substrates. It was also necessary to be able to list several anatomical sites of infusion in the same treatment within the model. The quantities of infused substrates were input in their original form, i.e. in $g.d^{-1}$, $mol.d^{-1}$ or $MJ.d^{-1}$. They were then homogenised and expressed in $kJ.h^{-1}$ of metabolisable energy or $g.h^{-1}$ nitrogen to take into account the nutritional contributions of the infused substrates.

The description of the methods in the publication varied from either referencing the author of the method and/or modifications to methods. It was decided to describe methods according to their ba-

sic principles, for example the colorimetric method [33], and enzymatic method [34] were used to describe the determination of ammonia-N.

The intermediate calculation steps between the elementary (blood/plasma flows and metabolite blood/plasma concentrations) and the final results (net nutrient fluxes) were not reported consistently among publications. Any missing blood and plasma flows or metabolite concentrations, were calculated from other results reported in the paper and validated. Moreover, to ensure the traceability of the results, a category "reported" or "calculated" was set up to accompany any flux result. In cases where the number of animals was missing, [35], it was decided to take the number of animals described in the Materials and Methods section of the publication, even when a publication reported results from several experiments with an apparently unequal number of animals per experiment. The results were reported using a variety of units, therefore it was decided to standardise them between publications using a molar and hourly basis of expression. When the standard errors were reported individually for each treatment, it was decided to calculate a mean standard error term for all the treatments.

3.1.2. Validation of the data within publications

One of the objectives of the data gathering was to allow the establishment of quantitative relationships between variables. Therefore, the first priority was to certify the coherence between the results reported within publications. An attempt was made to check that all variables from a given publication were coherent and referred to the same group of animals. First, all publications were systematically screened and such a screening had to be carried out before the input itself. Second, this screening

was defined as the cross-calculation of reported results from one to another (e.g. the re-calculation of net fluxes from concentrations and blood flows reported in the publication). Third, publications that showed large discrepancies between back-calculated and reported results were examined systematically to attempt to identify whether those discrepancies had a mathematical origin (rounding-up uncertainties, average of fractions, . . .) or a methodological origin (unequal number of observations within each average result reported). Furthermore, the uncertainty of measurement varied as a function of organs and tissues and metabolites. Therefore, the limits of rejection of the results were fixed. For example, for glucose net fluxes, the maximum tolerated discrepancy between the calculated results and those reported in the publication was arbitrarily set at ± 10% in the portal drained viscera. In comparison, the significant differences between treatments in this organ were about 20% ($P = 0.05$). Depending on these outcomes, a decision to utilise or reject the publication for inclusion into Flora was made.

### 3.1.3. Description of the dietary and nutritional conditions

The description of the nature of the feeds and diets, their chemical composition (CC) and nutritional value (NV) as well as the feed intake were strictly structured and defined to comply with the fourth specification of Flora. In order to thoroughly describe the nature of feeds and diets, it was decided to follow the INRA classification used in the feed tables [36]. First, all the information on the CC and NV value available in the publications was included. To correctly evaluate the intake, it was necessary to have at least the average amounts of dry matter intake per animal and per day. The unavailability of this information led to the rejection of the publication.

The most important parameters of CC and NV, namely gross energy, metabolisable energy, total nitrogen, digestible nitrogen, crude fibre, and organic matter digestibility, were included from each publication. If any of these values were missing, a procedure was used to estimate it (Fig. 1). To ensure the traceability of the estimation of the CC and NV values in Flora, it was decided to add a criterion to indicate whether data were reported or estimated. The validity of the method was established by estimating values which were otherwise known in the publication and by comparing the estimated and the known values. For some publications, the overall procedure of estimating the CC and NV was found to be time consuming, and therefore, this procedure was only carried out once it was decided to accept the publication into the database based on its results.

Following this, a final list of the necessary items and relevant essential data that was to be included in FLORA, was set up (Tab. I).

3.2. Development of the conceptual database model

The main entity of the data model was set to be the group of animals as already defined in Section 2.3. The first part of the model described the publication, its complete reference, its authors as well as the identification of the persons in charge of the input of data into Excel and their Research Service. It also described the publication, the experiments, the groups of animals and the experimental treatments and the relationships between them (Fig. 2a). The publication and its author were modelled by creating the two entities PUBLICATION and AUTHOR. The identification of the scientific operator in charge of the input of the data into Excel was described by an entity PERSON and two relationships called "is input by" and "belongs to"
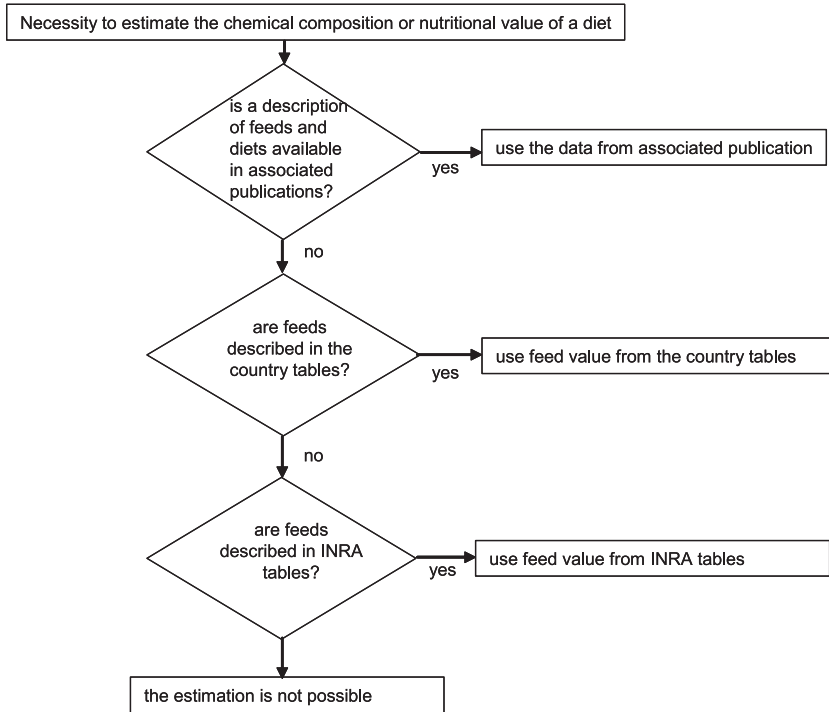
Figure 1. Procedure of estimation of the chemical composition (CC) and nutritional value (NV) of feed and diets. For mixed diets, CC and NV are calculated from the centesimal composition and the corresponding values of the simple feeds.

were created between PUBLICATION and PERSON and between RESEARCH SERVICE and PERSON, respectively. Furthermore, the entities EXPERIMENT, GROUP OF ANIMALS and TREATMENTS were created. Three relationships ("corresponds to") were created between PUBLICATION and EXPERIMENT, between EXPERIMENT and GROUP OF ANIMALS, and between GROUP OF ANIMALS and TREATMENT.

The second part of the model described the dietary characteristics and allowances and the ingredient composition of the diets, the chemical composition and the nutritional value of the feeds and diets, as well as daily intake (Fig. 2b). Thus two entities FEED and DIET were created, and linked together ("has an ingredient composition"). The chemical composition and the nutritional value of feeds and diets were described by a single entity FEED/DIET CHARACTERISTIC and two relationships ("has as chemical composition"). Finally, intake, expressed either on a dry matter basis, or in MJ of metabolisable energy or nitrogen per day, was linked to DIET and FEED CHARACTERISTIC by a relationship "is distributed at a given quantity".

The third part of the model described the treatments applied by way of substrate infusions (Fig. 2c). The potential mixture of substrates was modelled by an entity SUBSTRATE MIXTURE. The relationship "is composed of" was created between SUBSTRATE MIXTURE and SUBSTRATE (which gathered substrates such

Table I. Identification of the different items and corresponding essential data to extract from the publications.

Item	Essential data
Reference	- Title - Year of publication - Journal - Volume - Pages - Possible reference to associate publication(s)
Animals	- Species, breed, age, sex - Total number of animals per group - Physiological state
Surgical preparation	- Anatomical location of catheters in the body of the animal
Experimental treatment	- Nature of the treatment - Number of animals involved per treatment - average animal weight - Duration of the treatment - Type of treatment (nutritional, physiological stage, . . .)
Feeding, feed	- Category (concentrate, forage, . . .) - Mode of conservation – nature (hay, silage, . . .) - Vegetal family – product (grass, leguminous, . . .) - Vegetal species (Bermud grass, . . .) - Mode of preparation (pellets, . . .)
Feeding, diet	- Name of the diet - Type of diet (forage alone, . . .) - % of dry matter of the concentrate - Composition of the diet in the feeds (% dry matter)
Feeding, intake	- Diet number - Quantity parameter (dry matter intake $(g.d^{-1})$, . . .) - Value - "Reported/calculated" criteria - Reference for calculation
Feeding, composition of the feeds and diets	- Name of the parameter of chemical composition[a] - Value - "Reported/calculated/estimated" criteria - Reference for estimate - Reference for calculation
Feeding, substrate infusion	- Use of a mixture of substrates - Nature of the infused substrate - Site of infusion - Quantity parameter (metabolisable energy, nitrogen, . . .) - Amounts administered per day - Unit - Duration of the infusion - Nature of the diluant (water, . . .) - PH - Osmolarity

Table I. Continued.

Item	Essential data
Experimental period	- Number of days of measurement per treatment - Number of days between 2 successive treatments - number of days of recovery after surgery - Number of days of adaptation to the diet
Blood sampling	- Mode of sampling (continuous or spot sampling) - Number of samples collected per day and time interval of collection - Volume of blood collected per day
Methods of measurement of blood flow	- Principle of the method - Site of measurement - Criteria "blood/plasma"
Methods of determination of blood nutrients	- Principle of the method - Criteria "blood/plasma" - Products used for anticoagulation, deproteinisation, … - Conditions of refrigeration and conservation of samples type of sample (individual or pooled)
Statistics	- Experimental design - Analysis of results
Results on rumen parameters	- Name of the nutrient - Time of measurement - Type of result (concentration, …) - Value - Unit of the result - Number of animals - Criteria "reported/calculated"
Results on blood flow	- Name of the nutrient - Time of measurement - Value and name of the vessel - Criteria "blood/plasma" - Criteria "reported/calculated" - Number of animals - Reference for time - Reference for calculation
Results on blood fluxes	- Name of the nutrient - Time of measurement - Name of the vessel or organ or tissue - Type of result (concentration, difference of concentration, net flux, extraction rate) - Value of the result - Unit - Criteria "blood/plasma" - Criteria "reported/calculated" - Number of animals - Reference for time - Reference for calculation

Table I. Continued.

| Item	Essential data
Errors on blood flows	- Anatomical site of measurement - Type of result (average standard error, error term calculated for all treatments) - Criteria "blood/plasma" - Criteria "reported/calculated" - Value - Type of error (SEM, average SE)
Errors on blood fluxes	- Name of the nutrient - Type of result (concentration, . . .) - Name of the vessel or organ or tissue - Criteria "blood/plasma" - Criteria "reported/calculated" - Unit - Type of error (SEM, average SE)

[a]More explanations are given in the text.

as glucose, amino acids, . . .). The infused quantities were described by an entity PARAMETER OF QUANTITY and the relationship "corresponds to quantities" linking SUBSTRATE and PARAMETER OF QUANTITY. Finally, the characteristics of the infused solution were described by an entity INFUSED SOLUTION and the relationship "is infused as" which linked SUBSTRATE MIXTURE and INFUSED SOLUTION.

The fourth part of the model described the methods which were used for blood flow measurements and the analytical determination methods of the nutrient concentrations in blood, plasma and rumen fluid (Fig. 2d). Methods of blood flow measurements were described by the use of an entity ANATOMICAL SITE which represented in the present case the blood vessel and a relationship "was determined according to a method for blood flow measurement" which linked GROUP OF ANIMALS and ANATOMICAL SITE. An entity NUTRIENT was created which identified all the blood and plasma nutrients. Next, a relationship "was analysed according to a method" was established between GROUP OF ANIMALS and NU-

TRIENT. Finally, the chemical analysis of nutrients in rumen fluid was described by the entity RUMEN PARAMETER and the relationship "was analysed according to a method" between GROUP OF ANIMALS and RUMEN PARAMETER.

The fifth part of the model was created to describe the results of blood/ plasma flow and of nutrient concentrations in blood/plasma and rumen fluid (Fig. 2e). A relationship "measuring blood flow" was established between TREATMENT, and ANATOMICAL SITE. To model the results of blood and plasma nutrient fluxes, a new entity TYPE OF RESULT was created to gather all the types of results such as concentrations, concentration differences, net fluxes and fractional extraction. Then, a relationship "had a result on blood or plasma" was constructed to link TREATMENT, NUTRIENT and TYPE OF RESULT. Finally, a relationship "analysed in rumen fluid" was also created between TREATMENT and RUMEN PARAMETER.

The sixth part of the model was used to depict the other miscellaneous information which was present in the publications (Fig. 2f). An entity INFORMATION was
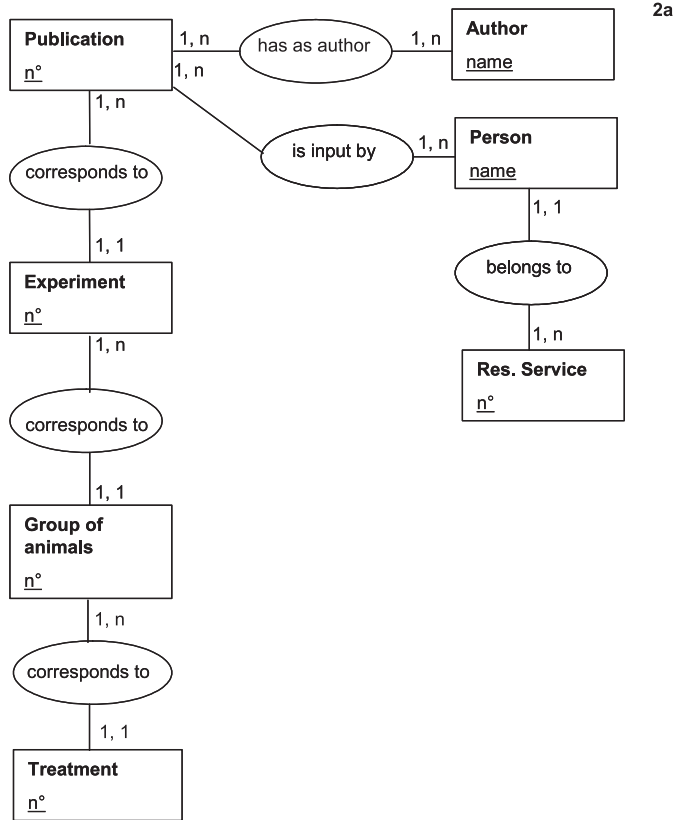
Figure 2. Representation of the conceptual data model for the bibliographic database on net blood nutrient fluxes: (**a**) first part of the model describing the reference of the publication and the authors as well as the person in charge of inputting the data, research service, publication, experiment, group of animals and experimental treatments; (**b**) second part of the model describing the feeds and diets characteristics and intake; (**c**) third part of the model describing the nature of the substrate mixture and substrates, the characteristics of the infused solution as well as the quantities infused; (**d**) fourth part of the model describing the methods of blood flow measurement as well ass blood determination; (**e**) fifth part of the model describing the measures of blood flow as well as the analyses of nutrients in blood, plasma and rumen fluids; (**f**) sixth part of the model describing the information on the experimental period, the catheter implantation, the blood sampling, the statistical methods as well as the daily meals. * Illustrated several times in the conceptus model.

created to gather information on the experimental period, catheter implantation, blood sampling, statistical methods, and the description of daily meals. A relationship "has information on" was created to link GROUP OF ANIMALS and INFORMATION.

3.3. Development of the relational and physical database

Entities and relationships were translated into computer tables in accordance with the MERISE method, [7]. Compared to the data model, no significant changes
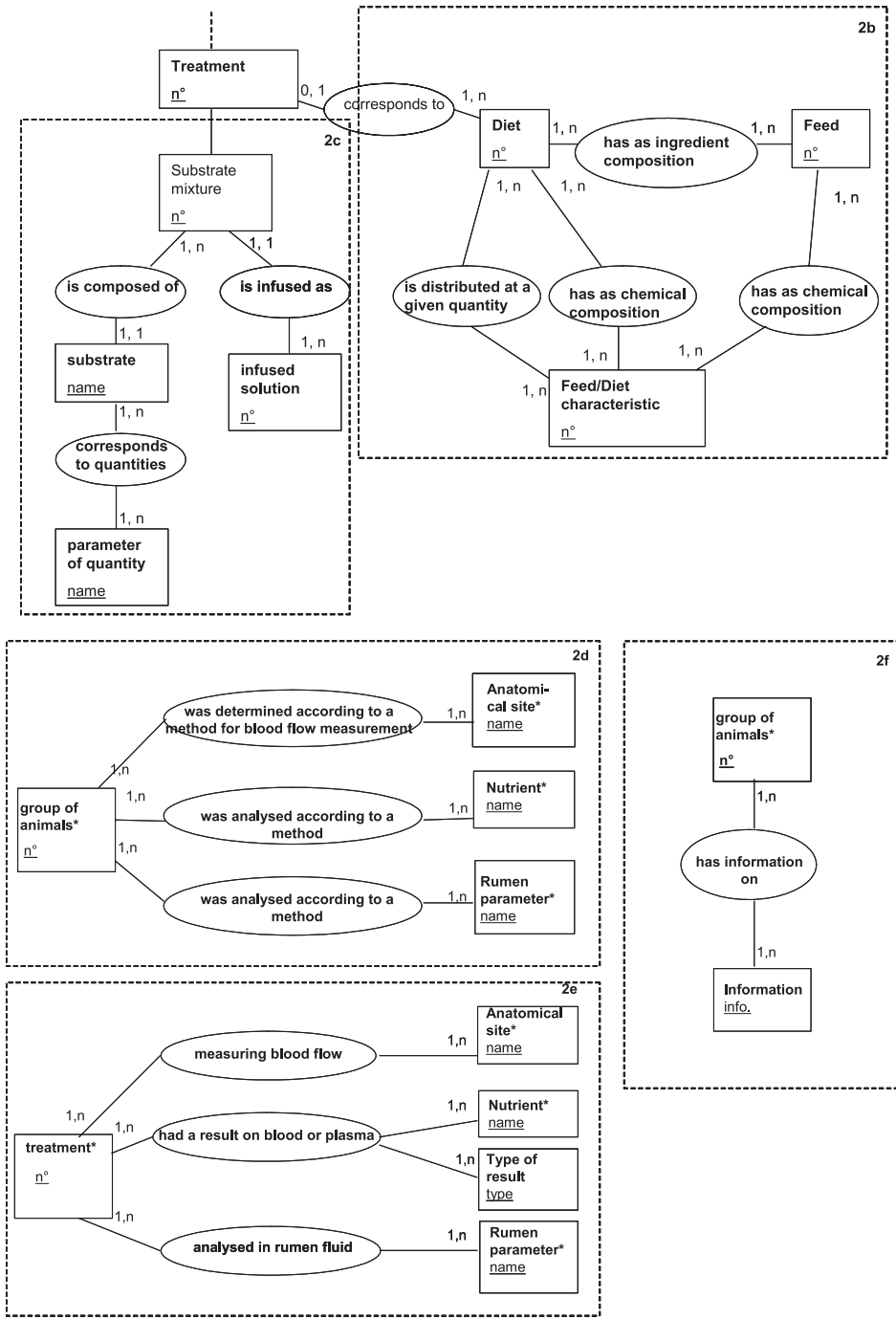
Figure 2. Continued.

and few new data groupings were carried out at this stage.

On the whole, 93 tables were created under Access [21] which were separated into reference tables ($n = 65$) and data tables ($n = 28$).

3.4. Additional tools

Data validation was implemented at the publication level. Its purpose was the calculation of the net nutrient fluxes from the blood flow and the nutrient concentration results (or vice-versa). It was developed under Excel [20] with separate sheets for each nutrient. The coherence of the results was evaluated by calculating the relative discrepancy between the back-calculated results and the reported ones.

Data input in Flora was created with Excel [20] in order to facilitate the data entry, checking and loading in Access tables [21]. Each publication had its own Excel file which was composed of 35 pairs of worksheets. Each pair of worksheet was devoted to a particular category of information, e.g. the treatment, diet, chemical composition of the diet... In each pair, the first worksheet was a "working worksheet" used for data input while the second one was used for the complete identification of the information, for example "service + publication" that would be transferred onto Access [21]. Drop down lists were connected to reference tables (last worksheet of the Excel file) in order to facilitate data entry to allow a complete and accurate description of feeds formalised as in [36] and to perform automated conversions of units.
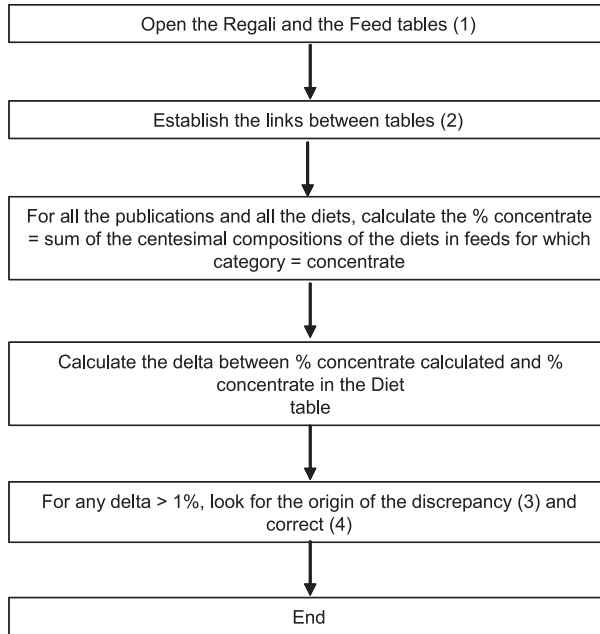
3.5. Data management

Data were managed in such a way that it was possible to check and verify that the data input onto Excel [20] and loaded onto Flora were correct, that there were no missing nor "double" data and that various information was coherent. The accuracy of the results of nutrient fluxes was checked in each publication by calculating the sum of the values both in the Excel file and the Access table and by comparing these 2 sums. Any missing or "double" data were detected by crossing the table of results with an identifier "service + publication + group of animals + treatment" corresponding to all the existing observations. Lastly, the search for coherence was done by recalculating the data from more elementary ones (e.g. the procedure of recalculating the percentage of concentrate of diets called upon the composition of diets (expressed in percent) and the description of feeds, forages or concentrates, Fig. 3).

Data retrieval from tables were done utilising queries by entering different parameters. For example, the parameters for a research on the results of glucose net blood fluxes in sheep at the level of portal drained vicera were as follows:" nutrient = glucose, type of result = net flux, compartment of measurement = blood, specie = ovine, site = portal drained vicera".

Two types of cross calculation were set up. The first one allowed to calculate the results of blood flow, plasma flow and hematocrite when 2 of these 3 results were known and the 3rd one was lacking. A second type of calculation made it possible to determine blood flow in the hepatic vein, hepatic artery and/or portal vein when 2 of these 3 results were known and the 3rd one was lacking (generally the flows in the hepatic artery were missing in the publications).

The description of the database made it possible to obtain elementary statistics (average, minimum, maximum, frequency) on the animals (species, physiological stage, weight), the diets, the chemical composition of the diets, the intake, the blood flow, the nutrient fluxes, the methods of measurement and analysis.

```
┌─────────────────────────────────────────────────────────┐
│          Open the Regali and the Feed tables (1)          │
└─────────────────────────────────────────────────────────┘
                            │
                            ▼
┌─────────────────────────────────────────────────────────┐
│             Establish the links between tables (2)        │
└─────────────────────────────────────────────────────────┘
                            │
                            ▼
┌─────────────────────────────────────────────────────────┐
│  For all the publications and all the diets, calculate the % concentrate │
│  = sum of the centesimal compositions of the diets in feeds for which    │
│                      category = concentrate              │
└─────────────────────────────────────────────────────────┘
                            │
                            ▼
┌─────────────────────────────────────────────────────────┐
│   Calculate the delta between % concentrate calculated and %  │
│                  concentrate in the Diet                  │
│                           table                           │
└─────────────────────────────────────────────────────────┘
                            │
                            ▼
┌─────────────────────────────────────────────────────────┐
│  For any delta > 1%, look for the origin of the discrepancy (3) and │
│                        correct (4)                        │
└─────────────────────────────────────────────────────────┘
                            │
                            ▼
┌─────────────────────────────────────────────────────────┐
│                           End                             │
└─────────────────────────────────────────────────────────┘
```

Figure 3. Procedure of checking of the percentage of concentrate in the diet: (1) Regali: relationship between diet and feeds + centesimal composition of the diet; Feed: description of the feed (name, category...); (2) the link between tables is made by identifiers: service No., publication No., group No., experiment No.; (3) return to the publication; (4) correction of the centesimal composition (Regali table) or percentage of concentrate (Diet table).

The automation of the estimation of missing data was carried out by crossing the table relating diets to feeds with the INRA table of chemical composition of reference feeds [36]. An estimation was obtained for the multi ingredient diets by adding the values of the individual feeds. A control of the estimation was carried out by comparing these estimated values with published values.
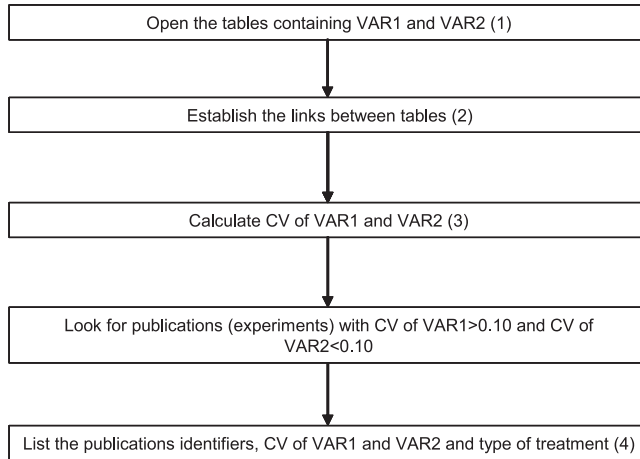
Several types of tables for meta-analyses were carried out to answer questions of research (see Sect. 4.4). They gathered identifiers (publication number, experiment number, group of animals number), information on the treatments, diets, animals, intake, the chemical composition and the feeding value of the diets, the values of blood flow and fluxes of nu-trients and the corresponding methods of measurement or analysis. The pre-coding treatments for meta-analyses were used to select publications corresponding to simultaneous variations on 2 or several variables X in intra publication (Fig. 4).

Data processing was carried out using the tool for creation of queries in Access [21].

4. DISCUSSION

The creation of the Flora database had one major objective, which was to gather and manage information from publications on multicatheterised ruminant animals, to ultimately enable the establishment of response laws of nutrient metabolism at

```
┌─────────────────────────────────────────────────────────────┐
│        Open the tables containing VAR1 and VAR2 (1)           │
└─────────────────────────────────────────────────────────────┘
                              │
                              ▼
┌─────────────────────────────────────────────────────────────┐
│             Establish the links between tables (2)            │
└─────────────────────────────────────────────────────────────┘
                              │
                              ▼
┌─────────────────────────────────────────────────────────────┐
│              Calculate CV of VAR1 and VAR2 (3)                │
└─────────────────────────────────────────────────────────────┘
                              │
                              ▼
┌─────────────────────────────────────────────────────────────┐
│   Look for publications (experiments) with CV of VAR1>0.10    │
│                  and CV of VAR2<0.10                          │
└─────────────────────────────────────────────────────────────┘
                              │
                              ▼
┌─────────────────────────────────────────────────────────────┐
│ List the publications identifiers, CV of VAR1 and VAR2 and    │
│            type of treatment (4)                              │
└─────────────────────────────────────────────────────────────┘
```

Figure 4. Procedure of pre-coding of meta-analysis to select publications with an important variation on VAR1 and a small or null variation on VAR2; (1) VAR1 and VAR2: explanatory variables; (2) the link between tables is made by identifiers: service No., publication No., group No., experiment No.; (3) CV: coefficient of variation; (4) type of treatment: nutritional strict, infusion, physiological stage.

the tissue level using meta-analytical approaches [19, 22, 37].

The choice was made to create a relational database rather than a spreadsheet for 3 main reasons. First, the expected data volumes were significant (about 30 000 lines for nutrient fluxes) and would be impossible to manage with a spreadsheet like Excel. Second, as already experienced by [11], a powerful tool of crossing of information was necessary to answer the numerous pre-planned or new questions of research which brought into play many types of data. Moreover, by crossing information, a relational database facilitates the detection of the missing or erroneous data and identifiers.

4.1. Development of Flora

For the development of the conceptual and operational database itself, three significant points are noteworthy. First, Merise proved to be a convenient method to model data from publications. From

this, 26 entities and 32 relationships were created to gather the different sources of information. The elaboration of the Flora database was facilitated by the existence of an experimental database on net nutrient fluxes which was already operational [9].

Flora was designed to store data from various types of experiments: nutritional experiments, experiments that studied the infusion of substrates and those that concern changes in the physiological state of the animal. Therefore, it was necessary to describe a significant number of experimental conditions (type of animals, feeding conditions, substrate infusion, blood sampling, methods of measurement and analyses...). Additional concepts were also introduced in Flora detailing the publications, authors, groups of animals, methods of measurement and modes of expression of the results. Furthermore, the identification of the data was completely re-examined to take into account the source of information (i.e. the name of the Research

Service, publication, experiment, group of animals and treatment).

The 28 data tables generated, were deduced from entities (e.g. DIET) and relationships. All the data tables were identified by the research service, publication, experiment and group of animals, to facilitate the relations between tables.

4.2. Compliance to specifications for the construction of the Flora database

Several specifications were defined for the construction of the Flora database.

In order to conform with the first specification, all necessary information was entered into the database, in order to describe in a precise manner all relevant items, including the authors, reference of the publication, animals, experimental treatments, feeds and diets, infused substrates, methods of measurement and analysis, results on rumen parameters concentrations, blood flow, and nutrient fluxes.

To satisfy the 2nd specification of easy data manipulation and retrieval, the choice was made to utilise a relational data base rather than a spreadsheet such as Excel [20]. Access was selected because it made it possible to store significant volumes (2 GO) [21].

A vital aspect for the entry of data into Flora was to avoid the risk of errors. Therefore, in order to meet the third specification as well as a wider specification of reliability, three points of control were used: the validation of data extracted from the publication, the verification of coherence of data within each publication and a final check that the data had been loaded accurately into Flora.

The fourth specification related to the detailed description of the dietary and nutritional conditions. First, in order to distinguish the various possible treatments, it was decided to introduce the concept of "type of treatment" into the database

and to describe in detail the treatments using "name of treatment". This description was very useful in particular when the treatments were calling upon interactions (e.g. intake x nature of the diet). Second, with regards to the nutritional characteristics, a minimum no of characteristics were decided. When these data were missing, estimations of these parameters were made, in order to have a full description of the chemical composition and of the nutritional value of feeds and diets. Reported and estimated values for publications in Flora agreed within ranges of uncertainty of $+3$ to $+5\%$ for crude protein, -1.9 to $+13.4\%$ for digestible nitrogen, -19.2% to $+12.5\%$ for crude fibre, -1.3 to -1.9% for digestible energy, and $+2.6$ to $+7.2\%$ for metabolisable energy e.g. [23, 24].

The fifth specification dealt with the precise description of the nutritional treatments when infusions were part of the experimental design of the publication. Flora was constructed to account for the mixtures of substrates and infusions in various anatomical sites. Moreover, it made it possible to input the data as they appeared in the publication hence avoiding tiresome conversions of units, and any risk associated with errors during such unit conversions.

Complying with the sixth requirement, the choice of classifying methods according to their principle proved convenient and feasible in most cases. This enabled methods to be fully detailed utilising all available information.

The traceability of information in agreement with good research practices stated in the seventh specification were also fulfilled. The traceability of the data was assured by adding a criterion "reported" or "calculated" or "estimated" as well as comments whenever it was judged necessary. Traceability was also ensured by adding "person" and "research service" in the database.

To take into account the within publication (or experiment) variability for subsequent meta-analyses [16, 19], (the eighth specification), the residual error term resulting from the variance analysis and the weighted average of the standard errors of the different treatments was considered as the most informative criteria and subsequently added to the database.

Limiting a unit choice (the ninth specification) reduced the number of unit conversions during the data input. Finally, the use of Excel [20] for the entry of all data reported above enabled easy data input and verification and subsequent transferal into the Access database for future management and retrieval (the tenth specification).

Finally, the rules of the management of Flora were established. The input of data into Excel [20] could be carried out by several different teams. However, the loading, control, update and extraction of the Flora database were under the responsibility of only a single person. This ensured that control measures at the input level were consistently met. However, easy access to the database for the various research teams involved in its construction was deemed vital. Therefore, data extraction was carried out upon request for specific types of animals and parameters in accordance with their research project.

4.3. Compliance to specifications for the data management

The main part of the procedures developed in the construction of Flora related to the control of the data, the cross calculations of data, the estimation of missing data (chemical composition of feeds and diets), the development of the tables of meta-analyses and pre-coding for the meta-analyses. All these procedures were possible thanks to the relational character of Flora which results of the use of the Merise method to construct it.

Data controls worked well. The limiting factors were the frequent returns to the publications to verify the known values, and the calculations utilised for the estimate of the chemical composition of diets, when these parameters were missing.

Currently, the procedures of cross calculations of data were carried out only for blood flow. They require careful management to consider all the possible combinations of data, in order to avoid mistakes in calculations or updates.

The automation of the estimation of missing data on diets was more efficient timewise compared to a manual estimation. However, it was often necessary to re-confirm the choices of feeds in the reference tables in order to get comparable results with the known values in the publication.

The procedure of development of tables for meta-analyses is complete to date. However, it required returning to the data control step when there were missing or repeated data.

The procedure of pre-coding of meta-analyses was a very interesting step for the Flora database. The two major difficulties were to thoroughly identify all the X variables which could have an influence on Y and to define for each X the thresholds of variation (i.e. the limit between non-significance and significance).

4.4. Uses of Flora

The Flora database has been used to establish quantitative response laws by meta-analysis on 1/ portal blood flow related to dry matter intake [38] and, 2/ the regulation of hepatic blood flow by feeding conditions in sheep [39] and 3/ net hepatic glucose release in ovines, as a function of dietary input [40]. It is currently being used to establish exhaustive response laws to relate the fluxes of energetic blood nutrients to feeding conditions in the ovine and bovine

(Bermingham, in preparation "Special Edition in Feed Sci and Tech").

In general, and compared to a database under Excel [20], the existence of a relational database under Access [21] facilitated the preliminary work of meta-analysis. First, it made possible the management of significant volumes of data (30 000 lines of results of nutrient blood fluxes). Second, it facilitated the selection of data (e.g. the selection of publications referring to energetic blood nutrients to be included in a meta-analysis). And finally, calculations of new variables from existing data were made possible (e.g. the calculation of the percentage of concentrate contained in the diets was made possible by connecting diets to feeds).

Since the creation of Flora, three teams have worked at the input of approximately 150 publications. For this work, the methods used for data input in Flora, the tools for the control of publications and the solutions chosen for the input of the measurement/determination methods and the characteristics of the infused substrates were all suitable.

5. CONCLUSIONS

Flora is a relational database. Therefore, it is possible to connect and cross, in an optimal manner, the different types of information and to consider all the necessary data processings. The most significant choices concerning Flora relate to estimating the missing data on feeds and diets.

Flora has been used successfully for quantitative and qualitative studies of the literature to establish first response laws by meta-analysis. It is currently being used in a larger way to determine response laws bringing into play many blood nutrients and various feeding conditions in sheep and bovines.

REFERENCES

[1] Sauvant D. La modélisation systémique en nutrition. Reprod Nutr Dev 1992, 32: 217–230.

[2] Majdoub L. Orientation propionique du profil fermentaire ruminal: conséquences sur le métabolisme splanchnique des nutriments énergétiques et sur la fourniture et l'utilisation du glucose par le muscle chez l'agneau recevant du fourrage vert, Thèse de Docteur de l'ENSAR, École Supérieure Agronomique de Rennes, 2002.

[3] Spedding CRW. General aspect of modelling and its application in livestock production. In: Kluwer AC pub (Ed), Modelling of livestock production systems. EEC, London, 1988, p 3–13.

[4] Sauvant D. Modelling in animal nutrition, story and interest for small ruminants. In: Eleventh seminar on the Sub-Network on sheep and goat nutrition, FAO-CIHEAM, 2005, p 129–131.

[5] Mann CC. Can meta-analysis make policy? Science 1994, 266: 960–962.

[6] Sauvant D. Le concept des lois de réponse multiples aux régimes, trait d'union entre les domaines techniques et économiques de l'élevage. in: INRA Institut de l'Elevage (Ed), Renc Rech Rumin, Paris, 1999, 6: 11–17.

[7] Lescourret F, Pérochon L, Coulon J, Faye B, Landais E. Modelling an information system using the MERISE method for agricultural

research: the example of a database for a system on performances in dairy cows. Agric Syst 1992, 38: 149–173.

[8] Ortigues I, Doreau M. Responses of the splanchnic tissues of ruminants to changes in intake: absorption of digestion end products, tissue mass, metabolic activity and implications to whole animal energy metabolism. Ann Zootech 1995, 44: 321–346.

[9] Vernet J, Majdoub L, Ortigues-Marty I. Relevance and elaboration of a base of experimental data of net blood nutrient fluxes across tissues and organs. Anim Res 2002, 51: 465–477.

[10] Seal CJ, Reynolds CK. Nutritional implications of gastrointestinal and liver metabolism in ruminants. Nutr Res Rev 1993, 6: 185–208.

[11] Ortigues I, Visseiche AL. Whole-body fuel selection in ruminants: nutrient supply and utilization by major tissues. Proc Nutr Soc 1995, 54: 235–251.

[12] Berthelot V, Pierzynowski SG, Sauvant D, Kristensen NB. Hepatic metabolism of propionate and methylmalonate in growing lambs. Livest Prod Sci 2002, 74: 33–43.

[13] Lescoat P, Sauvant D, Danfaer A. Quantitative aspects of blood and amino acid flows in cattle. Reprod Nutr Dev 1996, 36: 137–174.

[14] Isserty A, Ortigues I. Methods of analysis relating to blood flow measurement of the viscera and aorta in sheep. Reprod Nutr Dev 1994, 34: 399–413.

[15] Lescoat P, Sauvant D, Danfaer A. Quantitative aspects of protein fractional synthesis rates in ruminants. Reprod Nutr Dev 1997, 37: 493–515.

[16] Petitti D. Meta analysis and cost-effectiveness analysis, Methods for quantitative synthesis in medicine, Oxford University Press, New York, Oxford, 1994.

[17] Wang M, Bushman BJ. Integrating results through meta-analytic review using SAS software. SAS institute Inc., Cary, NC, USA. 1999.

[18] Rico-Gomez M, Faverdin P. La nutrition protéique modifie l'ingestion des vaches laitières: analyse bibliographique. In: Institut de l'Elevage (Ed), Renc Rech Rumin, INRA, Paris, 2001, 8: 285.

[19] St-Pierre NR. Invited review: Integrating quantitative findings from multiple studies using mixed model methodology. J Dairy Sci 2001, 84: 741–755.

[20] Microsoft Excel 2002 SP3. Microsoft Corporation, 1985–2001.

[21] Microsoft Access 2002. Microsoft Corporation, Version 2002, 1992–2001.

[22] Sauvant D, Schmidely P, Daudin J. Les méta-analyses des données expérimentales: applications en nutrition animale. INRA Prod Anim 2005, 18: 63–73.

[23] Goetsch AL, Ferell CL., Reynolds PJ. Effects of dietary maize level on net flux across splanchnic tissues of oxygen and nutrients in wethers consuming ad libitum different forages. Anim Sci 1995, 61: 43–55.

[24] Reynolds CK, Tyrell CL, Reynolds PJ. Effects of diet forage-to-concentrate ration and intake on energy metabolism in growing beef heifers: whole body energy and nitrogen balance and visceral heat production. J Nutr 1991, 121: 994–1003.

[25] Reynolds CK, Tyrell HF, Reynolds PJ. Effects of diet forage-to-concentrate ratio and intake on energy metabolism in growing beef heifers: net nutrient metabolism by visceral tissues. J Nutr 1991, 121: 1004–1015.

[26] Eisemann JH, Huntington GB, Catherman DR. Patterns of nutrient interchange and oxygen use among portal-drained viscera, liver, and hindquarters of beef steers from 235 to 525 kg body weight. J Anim Sci 1996, 74: 1812–1831.

[27] Thompson G, Manson W, Clarke P, Bell AW. Acute cold exposure and the metabolism of glucose and some of its precursors in the liver of the fed and fasted sheep. Quartely J Exp Physiol Sci 1978, 63: 189–199.

[28] Kristensen N, Danfaer A, Agergaard N. Diurnal pattern of ruminal concentrations and portal appearance rates of short-chain fatty acids in sheep fed a hay or a concentrate/straw diet in two meals daily. Acta Agric Scand Sect A Animal Sci 1996, 46: 227–238.

[29] Burrin DG, Ferrell CL, Eisemann JH, Britton RA. Level of nutrition and splanchnic metabolite flux in young lambs. J Anim Sci 1991, 69: 1082–91.

[30] Davenport RA. Data analysis for database design. Aust Comp J 1978, 10: 122–137.

[31] Lescourret F, Genest M, Barnouin J, Chassagne M, Faye B. Data Modeling for Database Design in Production and Health

Monitoring Systems for Dairy Herds. J Dairy Sci 1993, 76: 1053-1062.

[32] Tardieu H. La méthode Merise. Génie Logiciel 1986, 4: 25–29.

[33] Berthelot M. Violet d'aniline. Repert Chim Appl 1859, 1: 284.

[34] Kun E. Methods of enzymatic analysis. Bergmeyer, Verlag Chemie, Weinheim, and Academic Press, New York, 1974, p 1802–1806.

[35] Huntington GB, Prior RL. Digestion and absorption of nutrients by beef heifers fed a high concentrate diet. J Nutr 1983, 113: 2280–2288.

[36] INRA. Alimentation des bovins, ovins, caprins. INRA, Paris, 1988.

[37] Bangert-Drowns RL. Review of developments in meta-analytic method. Psychol Bull 1986, 99: 388-399.

[38] Vernet J, Lapierre H, Nozière P, Léger S, Sauvant D, Ortigues-Marty I. Prediction of blood nutrient supply to tissues of economical interest in Ruminants: a first step with the prediction of portal blood flow. In: Hill DRC, Barra V, Traore MK, Blaise Pascal University (Ed), First Open International Conference on modeling and simulation, OICMS, 2005, p 163-173.

[39] Vernet J, Nozière P, Léger S, Ortigues-Marty I. Regulation of hepatic blood flow by feeding conditions in sheep: a meta-analysis. In: Eleventh seminar of the sub-network on sheep and goat nutrition: advanced nutrition and feeding strategies to improve sheep and goat production, FAO-CIHEAM, Catania (It), 2005, p 132.

[40] Vernet J, Gely G, Léger S, Nozière P, Sauvant D, Ortigues-Marty I. Lois de réponse de l'émission nette de glucose par le foie chez l'ovin: méta-analyse. In: INRA Idl (éd), 11e Renc Rech Ruminants, Paris, 2004, p 264.