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■ A concise history of *Salmonella* spp. nomenclature

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■ Μια συνοπτική ανασκόπηση της ιστορία της ονοματολογίας του γένους *Salmonella*

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ABSTRACT. The history of typing strains of the genus *Salmonella* is a matter, perhaps, causing anxiety to those choosing to do research with this microorganism. The nomenclature and taxonomy of microorganisms of great Public Health importance, such as *Salmonella*, are causing proliferation of opinions and information representing various “schools of thought”. They produce a difficult to manage bulk of scientific information, eventually deterring inexperienced newcomers in this field of research. In overcoming this confusing proliferation of information, international bodies, having the responsibility of summarizing and officializing available knowledge, publish regularly “landmark decisions” on nomenclature and taxonomy. The present concise review of the history of *Salmonella* nomenclature aims in guiding the inexperienced researchers studying salmonellosis in animals and man toward the sources of accurate information.

Keywords: nomenclature, *Salmonella*, history

ΠΕΡΙΛΗΨΗ. Η ταυτοποίηση και ονοματολογία στελεχών του γένους *Salmonella* είναι ένα πολύπλοκο θέμα. Γενικότερα, η μεγάλη ερευνητική δραστηριότητα για τους μικροοργανισμούς που αφορούν τη δημόσια υγεία έχει ως αποτέλεσμα τη συσσώρευση επιστημονικών δεδομένων, η διαχείριση των οποίων συχνά είναι δύσκολη. Για το λόγο αυτό, οι διεθνείς οργανισμοί οι οποίοι ασχολούνται με την ταξινόμηση και ονοματολογία των μικροοργανισμών, σε τακτά χρονικά διαστήματα επαναπροσδιορίζουν τα κριτήρια με βάση τα οποία γίνεται η ταξινόμηση και η ονοματολογία τους. Στην παρούσα εργασία γίνεται μια σύντομη ανασκόπηση της ιστορικής διαδρομής της ονοματολογίας του γένους *Salmonella* με στόχο την κατανόηση της πληθώρας των πληροφοριών που αφορούν την ονοματολογία του συγκεκριμένου γένους.

Λέξεις ευρετηγίας: ονοματολογία, *Salmonella*, ταξινόμηση

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Introduction

The genus *Salmonella* is one of the most pathogenic members of the family of Enterobacteriaceae for man and animals. Many of its serotypes cause typhoid fever and severe diarrhea in man and a range of signs varying from subclinical to severe clinical enteric disease in animals (Acheson and Keusch 1997, Kuhns 2010). Salmonellosis is a foodborne infection, thus infected food producing animals are a health hazard requiring surveillance (SANCO 2009). Due to the microorganism's pathogenicity for man, it is systematically studied for more than 100 years by a plethora of scientists around the world, but, still today, many of its pathogenic characteristics are unknown.

The genetic base of *Salmonella* pathogenicity is better understood nowadays, since molecular methods are applied for studying the pathogen (Lim et al. 2005, Falush et al. 2006). These methods, in addition to associating the various isolated strains with the animal host or the type of clinical disease, have improved the existing knowledge on typing of the microorganism. They have not, however, given a definite answer to the many conflicting opinions continuously published on the nomenclature and taxonomy of the genus *Salmonella*. The lengthy and vigorous study of the genus *Salmonella* should have clarified these conflicting points, but, on the contrary, it further confuses those studying *Salmonella* spp. Thus, it is little help for the inexperienced scientists in appropriately reporting their research or clinical findings. The bulk of information, the many versions of "correct opinions", newer and older methods employed simultaneously, the choices of acclaimed agencies and laboratories and the difficulties of most of the others to follow their methodology, have been still confusing. Therefore, a concise review of this history could help them be encouraged when they are studying the microbe's pathogenicity and not its nomenclature or taxonomy. It could, also, provide them with a "friendly list" of sources to start their understanding of the confusing history of *Salmonella* spp. nomenclature. The search of the electronic databases on *Salmonella* and salmonellosis is another deterrent due to the bulk of information that is available, many times badly interpreted or referred to. Conflict and confusion for typing *Salmonella* spp. is the true history of an interesting and important microorganism.

Conflict and confusion for typing an important microorganism

The genus *Salmonella*, widely recognized as one group of microorganisms having worldwide importance, is the "star germ" for scientists arguing about bacterial nomenclature and taxonomy. The experts in microbiology propose, suggest, adopt and ignore opinions, perhaps, little thinking about those who will eventually have to use the names proposed, when reporting the possible causes of an observed clinical condition.

Organizations, such as the World Health Organization (WHO), the Centers for Disease Control (USA) and others (Internet sites, 2010), systematically inform scientists and the public about the conditions, pre-existing and newly described, caused by members of the genus *Salmonella*, but little information is given about the rules used in the typing of the cause.

Thus, older clinicians (medical doctors and veterinarians) report their work typing salmonella members in the way they are accustomed to, while their younger colleagues may find themselves struggling to understand what should be the official, thus the most appropriate, method of typing members of the genus *Salmonella*.

If their work is rejected for publication or severely criticized "for lack of current knowledge" about the nomenclature and taxonomy of *Salmonella* spp., they are discouraged to further pursue bacteriology, an already difficult field of microbiology. In the present short review, the aim is to give a reliable summary of the ongoing discussion of experts proposing names, species and schemes concerning the genus *Salmonella* from the point of view of an inexperienced scientist. It is hoped that the discussion will help young scientists and enthusiastic starting researchers understand how opinions influence science, if they are not indisputably proven as true or when they disregard the difficulties of many others around the world in adopting their suggestions. The history of the genus *Salmonella* is realistically reflecting the imposed scientific ambition, rather than the practical knowledge useful to everyone (clinicians and researchers).

Typing of a newly isolated microorganism

In 1884, T. Smith, a highly acclaimed veterinarian (Brown 1935) working as a young scientist for the

Bureau of Animal Industry, Department of Agriculture, USA isolated from the intestine of a dead pig a “bacterium”, which he thought was the cause of hog cholera (Smith 1894). Dr. Smith was at the time supervised by Dr. Salmon, also an acclaimed veterinarian of this time and a pioneer in animal disease control (Salmon 2010). The isolate of the two veterinarians was initially named “*Bacillus cholerae suis*”, but in 1900 it was elevated to the status of a genus by the French bacteriologist Liengieres with the name “*Salmonella*” (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934). The name, given in favor of Dr. Salmon by the chief of the Bureau of Animal Industry, is shadowing the contributions of the young scientist Dr. Smith, but, it is, also, reflecting the conflicts and ambitions of those working in pioneering fields of human and animal diseases. The genus *Salmonella* is a very large group of important microorganisms, clearly showing the reasons why a proliferation of opinions is characterizing all microorganisms of increased Public Health importance.

In 1900, the genus *Salmonella* Liengieres 1900 included a group of Gram (-) bacteria among which the best known are *Bacillus typhi-murium*, *Bacillus typhi*, *Bacterium paratyphi*, *Bacillus enteritidis*, *Bacillus cholerae-suis* (Brown 1935). However, the importance of this group of bacteria was increasing, as the reports with information relating them to a variety of clinical conditions, locations and animal species were increasing, coming from scientists working in the field of infectious diseases.

The 1st Congress of the International Society for Microbiology in 1930 was a landmark for the genus *Salmonella* Liengieres 1900. Shortly after this first congress, the nomenclature committee of the society formed a subcommittee having the responsibility to give answers to questions concerning the taxonomy and nomenclature of the genus *Salmonella* Liengieres 1900 (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934). Among the members of the subcommittee was Dr Kauffmann from Denmark, the “father” of the many thousands of salmonella serotypes.

The subcommittee published in 1934 the first official list with approved names for the genus *Salmonella* Liengieres 1900. The principle of the list was based on the “Kauffmann-White Scheme”, but it

was adhering as far as possible to the International Rules of Bacterial nomenclature. The Kauffmann-White Scheme is based on the presence or absence of specific antibodies against (O) and (H) antigens. (O) and (H) antigens were accidentally discovered in 1896 (Todar 2008, Euzeby 2010) and have been used since to group the genus *Salmonella*. The first officially reported list of groups of antigenically similar microorganisms using as differentiating characteristics the (O) and (H) antigens, divided the existing 44 species of *Salmonella* into five groups (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934).

Since the publishing of this list, microbiologists have attempted to successfully group strains isolated from various sources, developing eventually a complicated system of naming isolates. The system initially based on the concept of disease, animal species and area of first isolation, was giving new names, thus the status of species, to phenotypically and antigenically different isolates. Names, such as *Salmonella typhi*, *Salmonella typhi-murium*, *Salmonella enteritidis*, *Salmonella gallinarum*, *Salmonella abortus-ovis*, *Salmonella Cholerae-suis*, *Salmonella london*, *Salmonella panama* and many others included in the first official list of salmonella names, became common and are still familiar to field veterinarians and doctors (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934). Uninterrupted interest in the study of *Salmonella* serotyping increased further the number of species. The list of serotypes and species, becoming unmanageable for diagnosticians as early as in the 1940’s, required a change. Thus, opinions were published proposing a different thinking on the naming of microorganisms, in general, and members of the family of *Enterobacteriaceae* in particular (Borman et al. 1944).

The scientific opinion of the time on the taxonomy and nomenclature of the genus of *Salmonella* was that the multitude of names was “a deterrent to the progress in the field of medical bacteriology and, particularly, to the recognition of the importance of these organisms in public health” (Borman et al. 1944). This opinion of Borman et al. (1944) is still held true today, although Opinion 80, issued by the Judicial Commission of the International Committee on Systematic Prokaryotes (Tindal et al. 2005), has officially restricted the proliferation of species, however it is not

reducing the number of serotypes or restricting the number of species on the approved list of bacterial names (Euzéby 2010, Approved Lists of Bacterial Names 2010, Bacterial Nomenclature up-to-date: Approved List, Validation List 2010) to only two, as proposed by Opinion 80.

Borman et al. (1944), in criticizing the proliferation of names within the genus *Salmonella*, proposed the concept of the “few species” within which all other species and serotypes should be placed.

The concept of the few species

Borman et al (1944) proposed three species receiving the names *S. choleraesuis* for the type species, *Salmonella typhosa* (previously known as typhi) and *Salmonella kauffmannii*. The names were suggested only as the base for further knowledge on the concept of few species within the genus *Salmonella*, thus, they were, to the opinion of some, arbitrarily given (Kauffmann and Edwards 1952). *S. kauffmannii*, proposed in honor of the contributions of Dr. Kauffmann, was perhaps, an attempt to soothe the sharp criticism expressed on his contribution to the unmanageable proliferation of species within the genus *Salmonella* (Borman et al. 1944). *S. kauffmannii* was proposed to include all the known serotypes, the product of Dr Kauffmann’s scientific opinion (The Salmonella Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934).

Of course, proposals are useful, when scientific achievement is not overshadowing the need for practical thinking. Thus, practical “opinions” take time to mature, especially if a field is marked by the presence of scientists that are considered pioneers in their field, and *Salmonella* is still suffering the consequences of early pioneering opinion. However, the reduction in the number of species was becoming increasingly appealing, even to those working vigorously for many decades on the concept of the many species, like Dr. Kauffmann and his team. They, remaining firm in their concept of diagnostically serotyping salmonella, had early recognized the need of sub-grouping the genus of *Salmonella* and their thoughts were reflected in the first official list of species published in 1934 (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934). This list had divided *Salmonella* species and serotypes into five relative groups.

Decades later, Kauffmann and his team remained firm on the correctness of their method of serotyping salmonella, but they softened their approach to the concept of the three species (Kauffmann and Edwards 1952). In an attempt to appear as having their opinion on the matter, but not accepting earlier suggestions without objections, they proposed a biomedical scheme of grouping the recognized species of the genus *Salmonella* into three species, giving them the names *S. choleraesuis*, *Salmonella typhosa* and *Salmonella enterica*. This concept of dividing the genus was held valid to the 1970’s, when molecular typing became the new tool of research on strains isolated from clinical cases (Crosa et al. 1973). However, although molecular methods genetically relate isolates, they have not yet resolved the confusion created by the many *Salmonella* species and serotypes (Approved Lists of Bacterial Names 2010; Bacterial Nomenclature up-to-date: Approved List, Validation List, 2010, Euzéby 2010).

Thus, earlier opinions, held and based on less objective methods of typing the genus *Salmonella*, continued to come back, further confusing instead of helping the new reader of nomenclature and taxonomy for the genus. Between changes in the names of species, some proposed the division of the genus into sub-genuses or sub-genera (Kauffmann 2009). This grouping of the genus of *Salmonella* followed earlier concepts of grouping the known species and serotypes (The Salmonella Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934), eventually including the genus *Arizona*, too (Kauffmann 1960, Crosa et al. 1973, John Lindquist, 2010) having one species, the species *Arizona hunshawii* (Ewing 1969). The genus *Arizona* has some atypical similarities with the genus *Salmonella*, eventually becoming sub-genus III of the genus of *Salmonella* (Crosa et al. 1973, Rohde 1979, Tindal et al. 2005). The division of the genus into sub-genuses did not stop proliferation of new serotypes, but it had successfully stopped proliferation of species.

By 1973, when the first major molecular typing of *Salmonella* was attempted scientists were reporting results on the concept of three species, which had become *S. cholerae-suis*, *S. typhi* and *Salmonella enteritidis* (Ewing 1972). However, molecular typing of the genus showed that the species named during the history of studying the microorganism and its serotypes

could possibly form “one species” (Crosa et al. 1973). This discovery challenged for the first time both trends of thought; the early concept that each serotype was one species (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934) and the concept of few species (Borman et al. 1944).

The principle of the “one species”

The molecular typing of *Salmonellae* in the 1970's (Crosa et al. 1973) had started a new thinking on the nomenclature and taxonomy of this group of bacteria, but like all new methods, it was not yet fully challenged for its correctness. Thus, the first publishing of the Approved Bacterial Names, after molecular typing of the genus, continued to include five species. They were *S. arizone*, *S. choleraesuis*, *S. enteritidis*, *S. typhi* and *S. typhimurium* (Skerman et al. 1980). The observed diversity in opinion between those naming microorganisms and those seeking scientific proof of assumed differences show the existing differences between the attitudes of taxonomists, those setting the rules of nomenclature, those studying pathogenicity and those involved in clinical work (Crosa et al. 1973, Lim et al. 2005, Falush et al. 2006).

Since the first publishing of the Approved Bacterial Names (The *Salmonella* Subcommittee of the Nomenclature Committee of the International Society for Microbiology 1934), regular updates are published based on the opinions of the subcommittee of the Nomenclature Committee of the International Society of Microbiology. All proposals concerning changes in the taxonomy and nomenclature of bacteria are addressed to them for an official consideration. One such request for an opinion on acceptance of the principle of “one species” was made by LeMinor et al. (1982) many years after the first molecular typing of the genus (Crosa et al. 1973, Le Minor et al. 1982). They proposed to keep the name *S. choleraesuis* for the type species (Le Minor and Popoff, 1987), further dividing it into six sub-species named *S. choleraesuis* subsp *choleraesuis*, *S. choleraesuis* subsp *salamae*, *S. choleraesuis* subsp *arizone*, *S. choleraesuis* subsp *diarizonae*, *S. choleraesuis* subsp *hountanae* and *S. choleraesuis* subsp *bongori*. This proposal was also seeking an official opinion on the concept of sub-species within the genus. Their proposal was officially put in the Judicial Commission of the International

Committee of Systematic Bacteriology in 1987 together with a proposition to officially reduce the sub-genuses (I, IIa, IIIb, IV, V and VI) to the status of sub-species (I, IIa, IIIb, IV, V and VI). However, their request was rejected by the Judicial Commission on the assumption that such changes could eventually erase from memory the importance of serotype *S. typhi* (Wayne 1991). In addition, their proposal was invalidated as soon *S. choleraesuis* subsp *bongori* was described as a separate species taking the name *Salmonella bongori* (Reeves et al. 1989).

Almost simultaneously, the subcommittee of *Enterobacteriaceae*, which is a Member of the International Committee on Systematic Bacteriology, proposed, in the XIV International Symposium of Microbiology, the re-naming of the type species giving it the name *Salmonella enterica* (Penner 1988). The name “enterica” was proposed because it had not been previously used as a name of a serotype, thus it was less confusing compared to the name *S. choleraesuis*, appearing at times as species and at other times as a serotype. This proposal was, also, rejected, but the efforts to overcome judicial concerns continued (Euzeby 1999). This new proposal was seeking an exemption for serotype *S. typhi*, requesting its elevation to the status of species due to its clinical importance. The frequent requests of an opinion lead the Judicial Commission to publish the “Judicial Opinion 80”. With Opinion 80, the Commission agreed that from 2005 and thereafter, the name of the type strain (LT2^T) should be *Salmonella enterica* replacing the name *S. choleraesuis* (Euzeby 1999). The Commission issued explanations to bacteriologists on support of the change in the name and the taxonomy of *Salmonella* spp. (Tindal et al. 2005). With Opinion 80 the Commission accepted that the genus *Salmonella* had two species, *S. bongori* and *S. enterica*, and six sub-species belonging to *S. enterica* (Judicial Commission of the International Committee on Systematic Prokaryotes 2005). The name of each sub-species was formed using the name of the type species followed by the epithets *arizonae*, *diarizonae*, *enterica*, *hountanae*, *indica* and *salamae*. By that time, this opinion was officially published, a third species was identified and included in the approved list of 2005. It was named *Salmonella subterranean* (Shelobolina et al. 2004), which is today molecularly placed closer to *Escherichia hermannii* (Euzeby 2010), probably, forcing soon a new ruling and a new list of names.

The current types of *Salmonella* spp in published papers

Euzeby (2010), in his very informative and brief comment on the current standing of rules used in the nomenclature of *Salmonella* spp., clearly states that Judicial Opinion 80 (Judicial Commission of the International Committee on Systematic Prokaryotes 2005, Tindal et al. 2005) did not reject previous nomenclature. Thus, two systems of nomenclature are currently used. The one was validly published before 2005 and the other was published after 2005 and using the name *S. enterica* for the type strain. Thus, the current list of Approved Bacterial Names (Approved Lists of Bacterial Names 2010, Bacterial Nomenclature up-to-date: Approved List 2010), is the valid one. The most recent revision of it includes nine species and 14 sub-species. The species listed are *S. arizonae*, *S. bongori*, *S. choleraesuis*, *S. enterica*, *S. enteritidis*, *S. paratyphi*, *S. typhi* and *S. thyphimurium*. The sub-species are either *S. choleraesuis* followed by the epithets *arizonae*, *bongori*, *choleraesuis*, *diarizonae*, *houtenae*, *indica*, *salamae* or *S. enterica* followed by the above epithets.

The current list of Approved Bacterial Names appears as a compromise to all who historically influenced the names of the genus *Salmonella*. The number of species included on the list follows the early

concept of the few species (Borman et al. 1944). Although early molecular typing showed evidence that the concept of “one species” could be held valid to a point (Crosa et al. 1973), this concept cannot be scientifically sound for the present. Molecular methods have, like all methods, flaws because their results depend on the technique used and those working with the method (Crosa et al. 1973, Lim et al. 2005, Falush et al. 2006). Furthermore, before a very large data bank of genetic information is build and computerized, any new nomenclature received from molecular methods should be further examined.

Thus, they cannot be used reliably in taxonomy and nomenclature, but they should be considered important tools in relating isolates from different sources when epidemiological studies are conducted. In the case of *Salmonella* typing, they could eventually replace serotyping. In such a case, the complicated system of naming serotypes could be slowly replaced with an easier molecular system. If agencies and laboratories, such as the WHO, Centers of Disease Control and others, continue to serotype salmonella microorganisms rather than species or molecular types, the issue of *Salmonella* nomenclature and taxonomy will evolve in a confusing and complicated way, deterring the work of the inexperienced with the genus. ■

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