Proposal that *Mycobacterium massiliense* and *Mycobacterium bolletii* be united and reclassified as *Mycobacterium abscessus* subsp. *bolletii* comb. nov., designation of *Mycobacterium abscessus* subsp. *abscessus* subsp. nov. and emended description of *Mycobacterium abscessus*

Sylvia Cardoso Leao,¹ Enrico Tortoli,² Jean Paul Euzéby³ and Maria Jesus Garcia⁴

¹Departamento de Microbiologia, Imunologia e Parasitologia, Universidade Federal de São Paulo, São Paulo, Brazil
²Centro Regionale di Riferimento per la Diagnostica dei Micobatteri, Laboratorio di Microbiologia e Virologia, Ospedale di Careggi, Firenze, Italy
³Société de Bactériologie Systématique et Vétérinaire and École Nationale Vétérinaire de Toulouse, BP 87614, F-31076 Toulouse cedex 3, France
⁴Departamento de Medicina Preventiva, Facultad de Medicina, Universidad Autonoma de Madrid, Madrid, Spain

The names ‘*Mycobacterium abscessus* subsp. *abscessus*’ and ‘*Mycobacterium abscessus* subsp. *massiliense*’, proposed by Leao et al. (2009, *J Clin Microbiol* 47, 2691–2698), cannot be validly published. The purpose of this report is to provide a description in accordance with the Rules of the *Bacteriological Code* (1990 Revision). Moreover, the proposal of the name ‘*Mycobacterium abscessus* subsp. *massiliense*’ is contrary to Rule 38 and the correct name of this taxon, at the rank of subspecies, is *Mycobacterium abscessus* subsp. *bolletii* comb. nov. A description of *Mycobacterium abscessus* subsp. *abscessus* subsp. nov. and an emended description of *Mycobacterium abscessus* are also given.

The species *Mycobacterium bolletii* (Adékambi et al., 2006a) and *Mycobacterium massiliense* (Adékambi et al., 2004, 2006b) cannot be separated from *Mycobacterium abscessus* (Moore & Frerichs, 1953; Kusunoki & Ezaki, 1992) by phenotypic tests and results derived from genotypic studies, such as DNA–DNA hybridization and RFLP-16S rRNA, supporting the proposition that the three taxa actually represent a single species with internal variability. Consequently, Leao et al. (2009) proposed the union of *M. bolletii* and *M. massiliense*, and the recognition of two subspecies within *M. abscessus*: ‘*Mycobacterium abscessus* subsp. *abscessus*’ and ‘*Mycobacterium abscessus* subsp. *massiliense*’.

In contradiction with the *Bacteriological Code* (1990 Revision) (Lapage et al., 1992; De Vos & Trüper, 2000), Leao et al. (2009) do not provide protologues for the new subspecies (see Rule 27) and therefore the names ‘*Mycobacterium abscessus* subsp. *abscessus*’ and ‘*Mycobacterium abscessus* subsp. *massiliense*’ cannot be validly published by citation in a Validation List.

Moreover, the proposal of ‘*Mycobacterium abscessus* subsp. *massiliense*’ is not in accordance with the Rules:

- The name *Mycobacterium bolletii* was validly published on page 140 of the January 2006 issue of the *International Journal of Systematic and Evolutionary Microbiology*.
- The name *Mycobacterium massiliense* was validly published on page 2025 of the September 2006 issue of the *International Journal of Systematic and Evolutionary Microbiology*, by citation in Validation List 111.
- According to Rule 24b(2), the name *Mycobacterium bolletii* therefore has priority over the name *Mycobacterium massiliense*. Data presented by Leao et al. (2009) indicate that these taxa are heterotypic synonyms. According to Rule 38, if these taxa are considered to be one species, they must be united under the name *Mycobacterium bolletii*, and, according to Rule 50b, if that species is considered to be a subspecies of...
M. abscessus, then the name of the subspecies must be Mycobacterium abscessus subsp. bolletii.

According to Rule 27(2) (Lapage et al., 1992; De Vos & Trüper, 2000), a reference to a previous effectively published description is in accordance with the Rules. So, we propose the valid publication of the names Mycobacterium abscessus subsp. bolletii comb. nov. and Mycobacterium abscessus subsp. abscæsus comb. nov. with reference to the study by Leao et al. (2009) and the correction included in this report. The species description of M. abscessus is also emended to cover both M. abscessus subsp. bolletii and M. abscessus subsp. abscæsus.

The data from Leao et al. (2009), as well as other publications (Kim et al., 2008; Macheras et al., 2009; Zelazny et al., 2009; Leao et al., 2010), showed a clear internal variability at the phenotypic and genotypic levels in the species M. abscessus. Such a characteristic suggests strongly that possible future descriptions of other subspecies in the species M. abscessus cannot be discounted.

**Emended description of Mycobacterium abscessus (Moore and Frerichs 1953) Kusunoki and Ezaki 1992**

*Mycobacterium abscessus* (abs.ces’ sus. L. gen. n. abscessus of an abscess).

The following properties are displayed in addition to those listed in the species description by Kusunoki & Ezaki (1992): alkaline phosphatase, pyrrolidonyl arylamidase and tolerance to 5% (w/v) NaCl are present. Colonies can be smooth and grow on 5% sheep blood agar. Several enzymic activities show within-species variability, such as urease, catalase at 68 °C, β-glucosidase and β-galactosidase. More stable phenotypic characteristics are absence of growth at ≥42 °C, negative result for nitrate reduction and negative result for utilization of sodium citrate. The mycolic acid pattern, displayed by using HPLC, is very similar to those of other related species, showing only minor differences and low discriminatory power (Leao et al., 2009). *M. abscessus* has a single ribosomal operon (*rrn* operon) per genome (Domenech et al., 1994; Leao et al., 2009). This species has two characteristic PRA-*hsp65* patterns: type 1, showing *Bst*EII (bp) (235, 210) and *Had*II (bp) (145, 70, 60, 50), and type 2 with *Bst*EII (bp) (235, 210) and *Had*II (bp) (200, 70, 60, 50). The trait of 711 bp of the *rpoB* and the trait of 401 bp of the *hsp65* nucleotide sequences shows up to 96.6 and 98.7% within-species similarity, respectively.


The type strain is Hauduroy L948^T (=TMC 1543^T =ATCC 19977^T =CCUG 20993^T =CIP 104536^T =DSM 44196^T =JCM 13569^T =NCTC 13031^T).

**Description of Mycobacterium abscessus subsp. bolletii (Adékambi et al. 2006a) Leao, Tortoli and Garcia, comb. nov.**

*Mycobacterium abscessus* subsp. bolletii (bol.let’i. N.L. gen. masc. n. bolletii of Bollet, to honour Claude Bollet, a famous clinical microbiologist and taxonomist).


The description is based on that given for *Mycobacterium bolletii* by Adékambi et al. (2006a), with the addition of the description given by Adekambi et al. (2006b) for *Mycobacterium massiliense*. As demonstrated by Leao et al. (2009), the characterization of a wide set of isolates clearly showed further within-subspecies variability in this subspecies. Thus, the phenotypic characteristics of this subspecies show variation between strains in relation to the characteristics indicated in the first description given by Adékambi et al. (2006a, b). For example, several strains are able to grow in 5% (w/v) NaCl and others reveal a positive result for urease. The variability is even stronger when comparing antimicrobial susceptibility: several strains are susceptible to clarithromycin, in contrast to the resistance described in the initial report. It is important to mention that discrepancies in susceptibility tests could be also ascribed to the method used. *M. abscessus* subsp. bolletii has the properties given in the emended description of *M. abscessus*. Additional features that distinguish this subspecies are the PRA-*hsp65* pattern and the *rpoB* and *hsp65* gene sequence differences. The PRA-*hsp65* pattern that characterizes *M. abscessus* subsp. bolletii is *M. abscessus* type 2. The trait of 711 bp of the *rpoB* and the trait of 401 bp of the *hsp65* nucleotide sequences from *M. abscessus* subsp. bolletii show up to 95.6 and 98.5% similarity, respectively, to *M. abscessus* subsp. abscæsus. This subspecies includes strains of the species *M. massiliense* described previously (Adékambi et al., 2004, 2006b).

The type strain is BD^T (=CCUG 50184^T =CIP 108541^T =JCM 15297^T).

**Description of Mycobacterium abscessus subsp. abscæsus (Moore and Frerichs 1953) Kusunoki and Ezaki 1992, subsp. nov.**

*Mycobacterium abscessus* subsp. abscæsus (abs.ces’sus. L. gen. n. abscæsus of an abscess).

Rule 40d (formerly Rule 46) of the *Bacteriological Code* (1990 Revision) (Lapage et al., 1992; De Vos & Trüper, 2000) states that 'the valid publication of a subspecific name which excludes the type of the species automatically creates another subspecies which includes the type and whose name bears the same specific and subspecific epithets as the name of the type'. As *M. bolletii* is transferred to the species *M. abscessus* (*Mycobacterium abscessus* subsp. bolletii comb. nov.), an automatic consequence of this rule is that the subspecies *Mycobacterium abscessus* subsp. abscæsus (Moore and Frerichs 1953) Kusunoki and Ezaki 1992 must be created.
At the time of writing, the Judicial Opinion about the request by Tindall & Garrity (2008) has not been published. Thus, the authorship of the subspecies *Mycobacterium abscessus* subsp. *abscessus* follows the current Rule 40d.

The description is as that given for *Mycobacterium abscessus* by Kubica et al. (1972) and Kusunoki & Ezaki (1992). *M. abscessus* subsp. *abscessus* has the properties given in the emended description of *M. abscessus*. Additional features that distinguish this subspecies are the PRA-*hsp65* pattern and the *rpoB* and *hsp65* gene sequence differences. *M. abscessus* subsp. *abscessus* shows the characteristic PRA-*hsp65* pattern *M. abscessus* type 1. The trait of 711 bp of the *rpoB* and the trait of 401 bp of the *hsp65* nucleotide sequences from *M. abscessus* subsp. *abscessus* show up to 95.6 and 98.5% similarity, respectively, to *M. abscessus* subsp. *bolletii*.

The type strain is Hauduroy L948T (=TMC 1543T = ATCC 19977T = CCUG 20993T = CIP 104536T = DSM 44196T = JCM 13569T = NCTC 13031T).

### References


http://ijs.sgmjournals.org
Dear Authors,

Please find enclosed a proof of your article for checking.

When reading through your proof, please check carefully authors' names, scientific data, data in tables, any mathematics and the accuracy of references. Please do not make any unnecessary changes at this stage. All necessary corrections should be marked on the proof at the place where the correction is to be made; please write the correction clearly in the margin (if in the text they may be overlooked).

Any queries that have arisen during preparation of your paper for publication are listed below and indicated on the proof. Please provide your answers when returning your proof.

Please return your proof by Fax (+44 (0)118 988 1834) within 2 days of receipt.

<table>
<thead>
<tr>
<th>Query no.</th>
<th>Query</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Author: “utilization of” has been added before “sodium citrate”. Please confirm whether this is correct.</td>
</tr>
<tr>
<td>2</td>
<td>Author: 2006a or b?</td>
</tr>
</tbody>
</table>
SGM adopts a new way of ordering offprints

As a result of declining offprint orders and feedback from many authors who tell us they have no use for their free offprints, SGM has decided to phase out our practice of sending 25 free offprints to all corresponding authors.

We are also changing the way in which offprints are ordered. **When the final version of this article has been authorized for printing, you will receive an email containing a link to the SGM Reprint Service.** You can forward this email to your co-authors if you wish, so that they can order their own offprints directly, or to your finance or purchasing department, if orders are placed centrally.

When you click on the link in the email, you will be taken to an order page to place your offprint order. Like most online ordering sites, it will be necessary to set up an account and provide a delivery address while placing your order, if you do not already have an account. Once an account and delivery address have been set up, these details will be stored by the system for use with future orders. Payments can be made by credit card, PayPal or purchase order.

For an initial period, authors will be provided with a discount code that will allow them to order 25 free offprints, as well as any additional offprints they wish to purchase. This code will be valid for 90 days, and applies only to the paper for which it was issued. As all offprint orders will be despatched by courier from now on, there will be a charge for postage and packing, even on orders that consist only of free offprints.

Review authors will receive a discount code for up to 100 free offprints, as at present.

**SUMMARY**

- You can create or update your account at any time at [http://sgm-reprints.charlesworth.com/](http://sgm-reprints.charlesworth.com/)
- You will be sent an email when the offprints of this paper are ready for ordering
- **You cannot order offprints of this paper before this email has been sent**, as your paper will not be in the system
- You will also receive a discount code that will allow you to order 25 free offprints (postage & packing applies)
- Offprints can be ordered at any time after publication, although the discount code is only valid for 90 days

The ordering details and discount code will be emailed to the author listed as the corresponding author on the journal’s manuscript submission system. If your paper has been published (the final version, not the publish-ahead-of-print version) but you have not received your discount code, email reprints@sgm.ac.uk quoting the journal, paper number and publication details.

If you have any questions or comments about the new offprint-ordering system, email reprints@sgm.ac.uk