OPERATIONAL AND SCIENTIFIC NOTES

NOMENCLATURE OF *BACILLUS THURINGIENSIS* WITH ABBREVIATIONS

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ABSTRACT. *Bacillus thuringiensis* is accepted by most authorities as the valid specific name for insect pathogens in the genus *Bacillus* with subterminal spores, accompanied by crystals of delta endotoxin, and with biochemical reactions identical to those of *Bacillus cereus*. The main subdivisions of *B. thuringiensis*, termed 'varieties,' are based on numbered H-(= flagellar) antigens. Within some varieties crystal antigens are used to create subgroups of a lower order ('crystal-types'). Within both varieties and crystal-types, pathogenicity patterns in available host species can be used to characterize categories of a further lower order, possibly best regarded as 'strains.' Within varieties, uncharacterized isolates should be recorded by accession numbers. No abbreviations of names have been standardized or adopted as trade names and, when first used in a scientific paper, each abbreviation should be defined by the full scientific names.

INTRODUCTION. In the past two decades, work on *Bacillus thuringiensis* has increased both exponentially and rapidly. The number of known isolates and attempts to classify them increased proportionally. Although characters were found that gave comparable results allowing a relatively clear subdivisional structure to be created within the species, some confusion still arose about the definition of subdivisions. In addition, there has been long standing controversy concerning the specific name. The objective of this paper is to present a consensus of current opinions and to offer an authoritative opinion of future trends in nomenclature and abbreviations of names. The recommended terms are illustrated in Table 1.

SPECIES NAME. The insect pathogen, *Bacillus thuringiensis*, and the soil bacterium, *Bacillus cereus*, are very closely related. They are normally distinguished morphologically by the presence of a proteinaceous, crystalline, parasporal body of delta endotoxin accompanying the spore at sporulation in *B. thuringiensis*. Subdivisions within each species are clearly recognizable by their flagellar "H"-antigens (de Barjac 1981). Very few H-antigens of the two species cross-react at meaningfully low concentrations. Occasionally an isolate of *B. thuringiensis* loses the ability to form the crystal. Such isolates can be distinguished from *B. cereus* by H-antigens. However, acrystalliferous variants of the *few* isolates of *B. thuringiensis* whose antigens cross-react with those of an isolate of *B. cereus* cannot be distinguished from the particular *B. cereus* isolate that cross-reacts. On these grounds some purists argue that *B. thuringiensis* is a subgroup of *B. cereus*. However, by bacteriological standards, the ability to distinguish the two species—even with loss of crystal formation—is good. Also, ecologically, *B. thuringiensis* is primarily an insect pathogen and *B. cereus* primarily a soil organism. Consequently most authorities regard them as separate species. This nomenclature is now so well estalished and so useful in practice that the validity of the name *B. thuringiensis* is likely to survive (Table 1). Even if taxonomists argue otherwise in future, the name would probably be retained as a *nomen conservandum*.

SUB-GROUPS OF *BACILLUS THURINGIENSIS*. At

Table 1. Nomenclature and name abbreviations using *Bacillus thuringiensis* var *israelensis* as an example.

Hierarchial status	Full name and best abbreviation	Definitive characters	Current numbers of each hierarchy
Genus	*Bacillus*	Morphology, biochemistry	—
Species	*thuringiensis* (*B.t.*)	Morphology	—
Variety (not subspecies)	*israelensis* (*B.t.i.*)	Flagellar antigen, H-14	27 varieties 24 H-types
Crystal serotype	*isr*	Crystal antigen	*ca.* 28
Strain	None: arbitrary numbers within varieties	Toxicity in groups of hosts	*ca.* 84
Isolate	e.g. HD-917* (de Barjac 1884**)	Isolation date, source and person who isolated it	900+
Mutant	e.g. HD-917/1	Antibiotic resistance, auxotrophy, pathogenicity, etc.	0 to 50+ per strain

* Accession number in the culture collection of H.T. Dulmage, USDA, Brownsville, Texas, USA.
** Accession number of same isolate in the collection of H. de Barjac, Pasteur Institute, Paris, France.

present, 24 H-antigen groups are recognized and a procedure has been published for their logical numbering (Burges et al. 1982). This involves checking, before publication, with Prof. H. de Barjac at the International Reference Centre, Pasteur Institute, Paris. A proposed new H-serotype is compared with other new types in the process of numbering to ensure that it is different.

Since the H-serotypes agree remarkably well with classifications based on patterns of esterases from vegetative cells (Norris and Burges 1965), crystal antigens (Krywienczyk, in Dulmage and cooperators 1981) and classical biochemistry (de Barjac 1981), they are undoubtedly the natural first order of subgroups (although susceptibilities to different bacteriophages cut indiscriminately across them). In the 8th edition of Bergey's Manual (Buchannon and Gibbons 1974) these subgroups were designated as "subspecies." This has not proved popular and is likely to be discontinued in favor of "varieties" in the next edition and in common usage. Thus the correct format for writing a first order subgroup (a variety) is, for example, *Bacillus thuringiensis* var *israelensis,* using italics for the varietal name. Varieties are mainly defined by the flagellar serotype which is designated by the serotype number, e.g., *B. thuringiensis* H-14. This number is sometimes used in place of the varietal name, a practice that should be limited to follow an initial use of the varietal name (Table 1).

Within many varieties, subgroups can be formed on the basis of crystal antigens. These have been designated by the first three letters of the varietal name e.g., *isr* (from *israelensis,* Table 1), or by the collector's code number for the isolate in which the new antigen was first found, in italics, e.g., *B. thuringiensis* var *kurstaki k-73* (Dulmage and co-operators 1981). These subgroups are best called "crystal types," not strains, because each subgroup contains isolates that can be distinguished as different from each other.

Within each variety and crystal type, there can be further subdivisions based on differences in pathogenicity to a variety of host species (Dulmage and cooperators 1981). At present it seems reasonable to regard member isolates within these "pathogenicity" groups as belonging to a "strain" (designated by an arbitrary number, Table 1), since they cannot be reliably distinguished between each other. Until characterized as belonging to such a numbered strain, a newly isolated organism is best termed an isolate within a named variety and designated by a culture accession number. As isolates are passed from collection to collection, they receive new accession numbers. To enable the initial source and subsequent history of an isolate to be traced, all known accession numbers attached to an isolate should be documented in publications (Table 1).

Jones et al. (1983) were able to distinguish available isolates in var *aizawi* (H-serotype 7) by bacteriophage typing, but studies with other varieties are needed before this can be regarded as a general phenomenon. Jarrett (1983) distinguished between numerous isolates in H-serotype 7 by the patterns of their extrachromosomal DNA elements (plasmids). However, plasmids are too easily lost and gained for this to be more than a limited short-term means of isolate recognition.

The advance of genetical studies with *B. thuringiensis* has resulted in the production of many mutants. These are normally designated by some form of number added to the number of each "wild" isolate (Table 1).

ABBREVIATIONS. Even though the name "*B. thuringiensis*" and those of its varieties are now so frequently used, standard abbreviations are never likely to be designated for them after the fashion of "EDTA" in chemistry and "DDT" in pest control. Abbreviations (Table 1), such as *B.t.,* should not, therefore, be used in the titles of publications. They should not be used in summaries and the text until defined by the full scientific name *viz., B. thuringiensis (B.t.).* Since none of the abbreviations is standard, they are inserted at the discretion of individual authors. "*B.t.*" for *B. thuringiensis,* "*B.t.i.*" or "*B.t.* H-14" for *B. thuringiensis* var *israelensis,* "*B.t.* H-3a3b" for *B. thuringiensis* var *kurstaki,* etc., are common and acceptable. "*B.t.i.*" should not be used to include isolates in general that are active against vectors, because some new isolates of other varieties also have proven active against vectors. "*B.t.t.*" is totally unacceptable as a generalization for isolates active against agricultural pests, because many varieties have this character.

Since the choice of abbreviation by different authors varies, an editor of a journal should choose arbitrarily between equally acceptable alternative abbreviations such as "*B.t.i.*" and "*B.t.* H-14" and, thereafter, standardize on the choice for consistency in the journal.

NAMES OF COMMERCIAL PRODUCTS. Many commercial names now describe specific products from specific manufacturers, e.g. Technar®. These should always have the first letter a capital and the variety involved, if known, should always be mentioned in a publication as a full name. Due to product variation, batch numbers also should be mentioned, a practice already found useful in literature reviewing. The only instance where an abbreviation, such as "*B.t.i.,*" could be regarded internationally as standard would be if it were adopted

as a registered trade name for a specific commercial product, which has not happened yet.

STANDARD PREPARATIONS. It is normal practice to bioassay both commercial and experimental products against internationally accepted standard products, which permits a description of potency in terms of international units. A succession of standard powders, IPS78, IPS80 and IPS82 has been produced for the *israelensis* variety, each powder consecutively replacing its predecessor. For other varieties, products numbered E61, HD-1, etc., have been used. These products form unique series, with numbers different from—and not to be confused with—numbers used for serotypes, strains or isolates (Table 1).

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LABORATORY OBSERVATIONS ON SOME FRESHWATER VERTEBRATES AND SEVERAL SALINE FISHES EXPOSED TO A MONOMOLECULAR ORGANIC SURFACE FILM (ISA-20E)

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Several mosquito species have been tested against the monomolecular organic surface film Arosurf 66-E_2 (ISA-20E) from the Sherex Chemical Company, to determine the percentage of kill (LD50 or LC90) of the film on these species, but work has just begun on non-target species associated with mosquito habitats. We evaluated the effects of this mono-oil on the fresh-water green tree frog, *Hyla cinerea* (Hylidae) and on the two fresh-water fishes, *Hypostomus plecostomus* Loricariidae and *Gambusia affinis* Baird and Girard (Poeciliidae). We also tested the following saline fish species with ISA-20E under aerated conditions: *Fundulus confluentus* Goode and Bean, *Fundulus grandis* Baird and Girard and *Cyprinodon variegatus* Lacepede (Cyprinodontidae), *Poecilia latipinna* Lesveur (Poeciliidae) and *Dormitator maculatus* Bloch (Eleotridae).

Early in September 1981, the three freshwater individuals were placed in an office aquarium containing conditioned tap water at varied room temperatures and photoperiods. After a week, the monomolecular surface film was added at an equivalent to 0.68 ml/m^2 to the 10 gal (45 liter) fish tank. For the next 6 months (September 1981–February 1982) the specimens were observed and fed. The mono-oil surface was maintained.

The green tree frog progressed normally from tadpole to adult and the two fish survived and developed normally.

Based on our observations of these three freshwater species, exposed to the mono-oil film for 6 mo, we determined that the ISA-20E had no detrimental effect.

Fish were collected from the salt marsh in unbaited barrel minnows traps and with dip nets. Salinity readings were taken and ranged from 20 to 44%. The fish were left overnight in the aquarium room in their own water.

The aquarium lab is an insulated, converted, 10 × 20 foot, storage building with a 12,000 BTU fully automated heat-cool air conditioner.