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TOOLS FOR PROTEIN SCIENCE

Collection of antimicrobial peptides database and its derivatives: Applications and beyond

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Abstract

Collection of antimicrobial peptides (CAMP), CAMPSign, and ClassAMP are open-access resources that have been developed to enhance research on antimicrobial peptides (AMPs). Comprehensive information on AMPs and machine learning-based predictive models are made available for users through these resources. As of date, $CAMP_{R3}$ has 10,247 sequences, 757 structures, and 114 family-specific signatures of AMPs along with associated tools for AMP sequence and structure analysis. CAMPSign uses family-specific sequence conservation, in the form of patterns and hidden Markov models for identification of AMPs. ClassAMP can be used to classify AMPs as antibacterial, antifungal, or antiviral based on sequence information. Here we describe CAMP and its derivatives and illustrate, with a few examples, the contribution of these online resources to the advancement of our current understanding of AMPs.

KEYWORDS

antimicrobial peptides, CAMP database, CAMPSign, ClassAMP, family-specific signatures, machine learning

1 | INTRODUCTION

Antimicrobial peptides (AMPs) are multifaceted host defense molecules that are produced by organisms ranging from microbes to mammals.[1] AMPs kill microbes via pleiotropic mechanisms of action, such as destruction of the microbial membrane and inhibition of macromolecule synthesis.[2–4] These molecules exhibit broad range antimicrobial activity, rapid killing kinetics, reduced toxicity, and reduced microbial resistance. Apart from their antimicrobial activity, a few AMPs also regulate physiological functions such as inflammation, angiogenesis, and wound healing.[5]

Identification of AMPs from natural sources in the 1980's stimulated research on their isolation and characterization. Further, the threat posed by antibiotic resistance accelerated research on AMPs. This resulted in a rapid increase in the number of identified AMPs, which in turn demanded efficient AMP data registration, organization, and retrieval methods. Responding to this need, a few databases on AMPs were developed.[6–14] However, these databases were limited to AMPs from a specific source, that is, either natural or synthetic, or from a particular source organism. To address this issue, we developed the collection of antimicrobial peptides (CAMP) database[15] in the year 2010. The manually curated sequence information of AMPs was further used to develop tools for AMP prediction and annotation. The databases and tools, that were developed by our group and updated over time, are discussed below.

2 | CAMP DATABASE

2.1 | $CAMP_{R1}$

CAMP was an open-access resource that provided information on sequences of natural as well as synthetic AMPs on a single platform. AMP sequence information obtained from the publicly available National Center for Biotechnology Information (NCBI) database was systematically categorized as (a) experimentally validated, (b) predicted, and (c) patents

based on the reference literature. Furthermore, information on the target organism, MIC values and hemolytic activity was manually annotated from literature. It was the first AMP database to have information on patented AMPs. The database also hosted machine learning (ML) based algorithms like random forests (RF), support vector machines (SVM), and discriminant analysis (DA) for AMP prediction.

2.2 | $CAMP_{R2}$

CAMP was expanded in 2014 to include information on structures of AMPs.[16] An additional feature of $CAMP_{R2}$ was the introduction of family information for sequences present in the database. AMPs belong to diverse families such as cathelicidins, defensins, temporins, and so forth, having characteristic sequence composition. The database had manually annotated information on 53 AMP families. This information was meticulously sourced from (a) UniProtKB,[17] (b) protein family databases such as Pfam,[18] InterPro,[19] and (c) literature databases such as PubMed. The database was updated with newly identified AMPs and the prediction algorithms were retrained using the updated sequence information.

2.3 | $CAMP_{R3}$

The inclusion of AMP family-specific signatures represented by patterns and hidden Markov models (HMMs) and the AMPs retrieved from online databases using these signatures, mainly constituted the third update of CAMP. Users can access $CAMP_{R3}$[20] for information such as family signatures, sequences, structures, activity profile, source, target organisms, hemolytic activity and links to external databases such as UniProtKB, PDB, PubMed, and NCBI Taxonomy for AMPs from eukaryotic and prokaryotic sources. Presently, $CAMP_{R3}$ holds information on 10,247 sequences, 757 structures, and 114 family-specific signatures of AMPs along with associated tools for AMP analysis. Thus, CAMP evolved from a simple repository of AMP sequences to a comprehensive database containing sequences, structures, and family signatures along with associated tools for AMP analysis. The evolution of the CAMP database from its inception to the present state is described in Table 1.

3 | ONLINE WEBSERVERS FOR ANALYSIS OF AMPs

3.1 | CAMPSign

AMPs belong to diverse families with conserved sequence composition and this can be leveraged to efficiently identify/predict AMPs from a large pool of sequences. CAMPSign is an open-access webserver that aids in identification of AMPs and their families using family-specific signatures represented by patterns and HMMs.[21] CAMPSign, presently can predict members of 45 AMP families.

3.2 | ClassAMP

While few AMPs exhibit broad-spectrum activity, many of them are target-specific. ClassAMP is an online prediction tool for classification of peptides as antibacterial, antifungal and/or antiviral using sequence-based features.[22] It employs ML algorithms such as SVM and RF for classification.

4 | RESOURCES AVAILABLE AT CAMP AND ITS DERIVATIVES

4.1 | Database search

The CAMP database can be searched to retrieve sequences, structures and signatures of AMPs. The database provides basic and advanced search options. Basic search feature enables keyword-based search for all fields or restricted to a specific field. The advanced search has a query builder by which users can combine multiple queries using logical AND or OR operators. Users can search the database using AMP name, sequence, source organism, target organism, activity, and so forth. Users can also query for AMP family members and retrieve family signatures in the form of patterns and HMMs.

4.2 | Data analysis tools available through CAMP

4.2.1 | AMP prediction

Users can input sequences and obtain a variety of information including the following: (a) Predict if the sequence/s is/are antimicrobial; (b) Predict whether antimicrobial regions are present within a protein; (c) Rationally design single-residue mutants

TABLE 1 Updates of the CAMP database

Database	Primary structure	3D structure	Family description	Patterns and HMMs
CAMP (2010)	4,020	–	–	–
$CAMP_{R2}$ (2014)	6,756	682	3,111 (53 families)	–
$CAMP_{R3}$ (2016)	10,247	757	5,241 (53 families)	114

Abbreviations: CAMP, collection of antimicrobial peptides; HMMs, hidden Markov models.

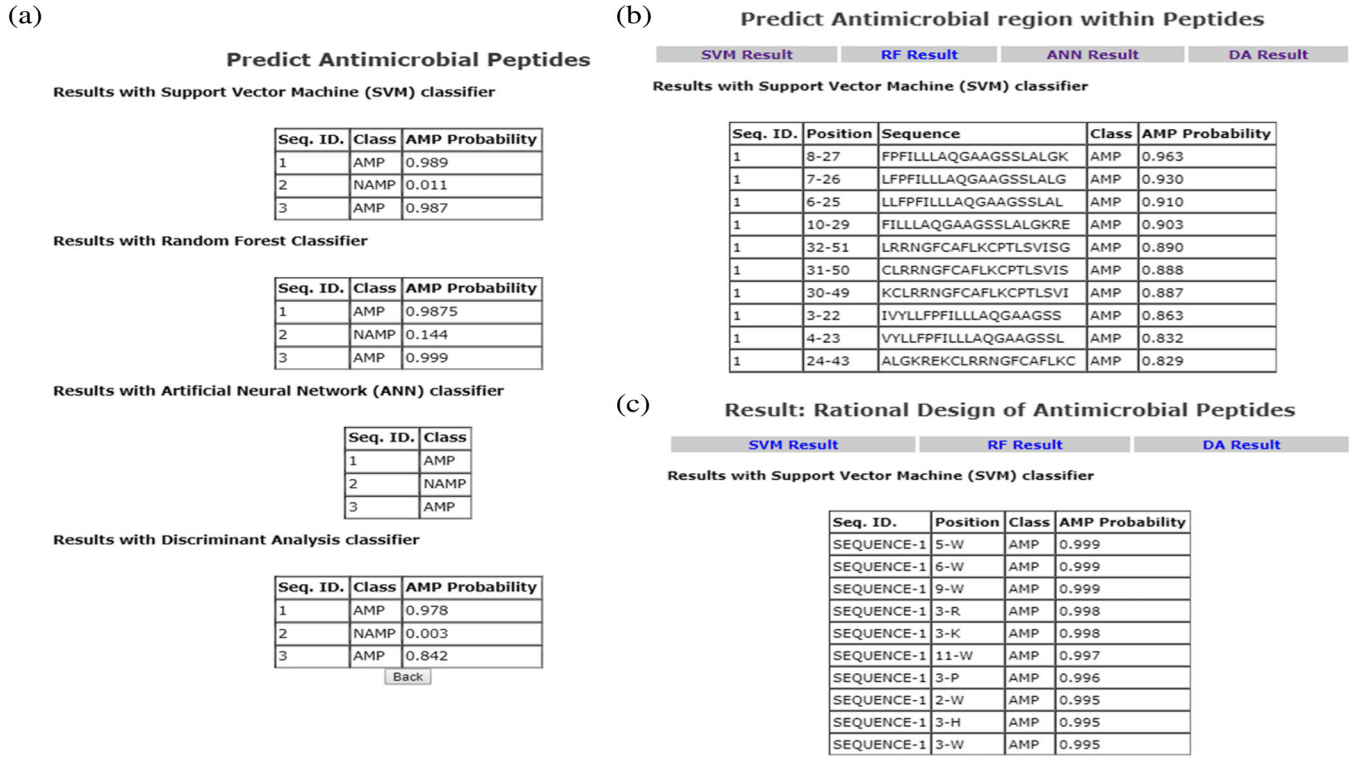

FIGURE 1 Snapshots of analysis reports from prediction modules available in CAMP. (a) Prediction of antimicrobial peptides based on various ML classifiers. (b) Prediction of antimicrobial regions within the input sequence. (c) Prediction of single-residue mutants with enhanced antimicrobial activity. A probability threshold of 0.5 was heuristically set to classify peptide as being antimicrobial. CAMP, collection of antimicrobial peptides; ML, machine learning

and predict the effect of substitutions on antimicrobial activity. The outcome of the prediction analysis is in the form of probability scores (Figure 1). The higher the score (max = 1), the greater is the likelihood of the sequence having antimicrobial activity. The prediction tools aid in the identification and rational design of novel AMPs.

4.2.2 | Basic local alignment search tool

Users can query peptides of interest and their homologs in the entire CAMP database or restricted to various subdatasets of CAMP, for example, structure, patent, experimentally validated, predicted, and predicted based on signature data sets using the basic local alignment search tool (BLAST) tool[23] available in CAMP.

4.2.3 | Links to third-party analysis tools

The CAMP database provides access to third-party tools for sequence and structure analysis such as Clustal Omega,[24] Vector Alignment Search Tool (VAST),[25] PRATT,[26] ScanProsite,[27] PHI-BLAST,[28] and jackhmmer[29] for increasing the data analysis options available to users.

4.3 | Predict AMPs based on family signatures

The CAMPSign webserver can be used to predict AMPs based on family-specific sequence conservation. Users can scan the sequence of interest against all or specific AMP family signatures comprising of patterns and HMMs. Results are generated in a tabular format and have information on the AMP family and number of patterns and HMMs that match user-defined sequence/s. Users can obtain a detailed view of the patterns and HMMs that match/align to the sequence through the hyperlinks in the results page (Figure 2).

4.4 | Predict AMPs based on target organisms

The ClassAMP webserver can be used to predict the propensity of a peptide to have antibacterial, antifungal, or antiviral properties based on sequence features trained using SVM or RF based algorithms. The sequences are predicted as antibacterial, antifungal, or antiviral and a probability score (0–1) is provided. The higher the probability score, the higher the likelihood of correct classification.

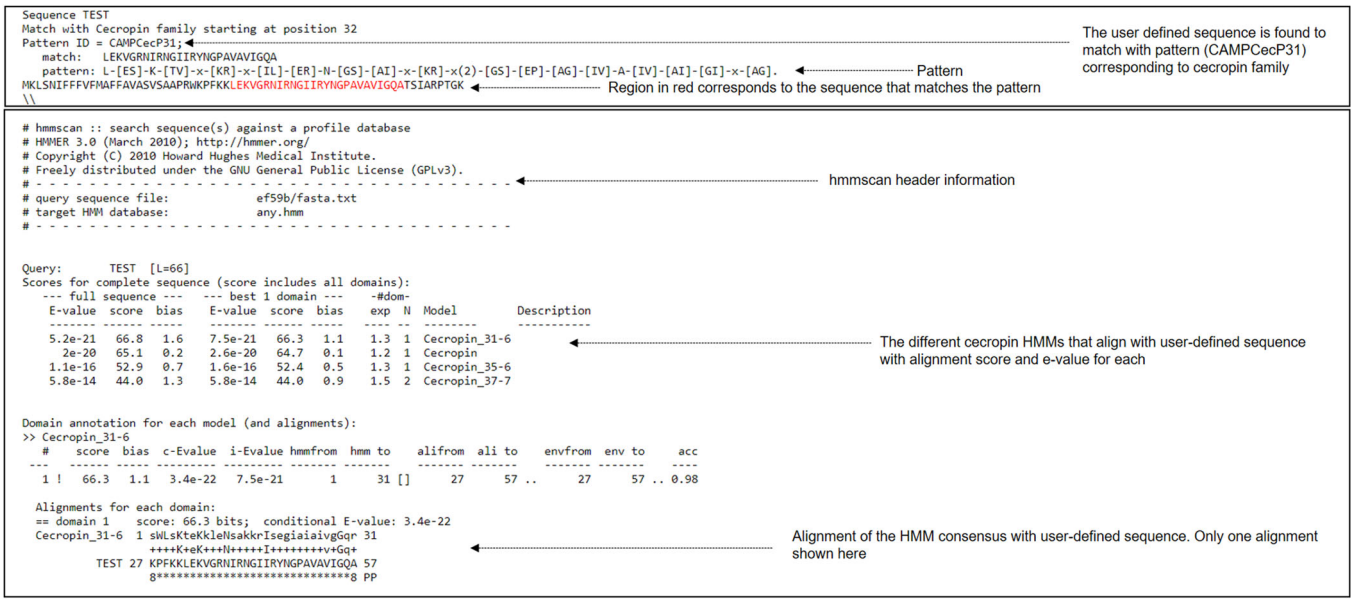

FIGURE 2 Screenshot and description of the analysis report of CAMPSign

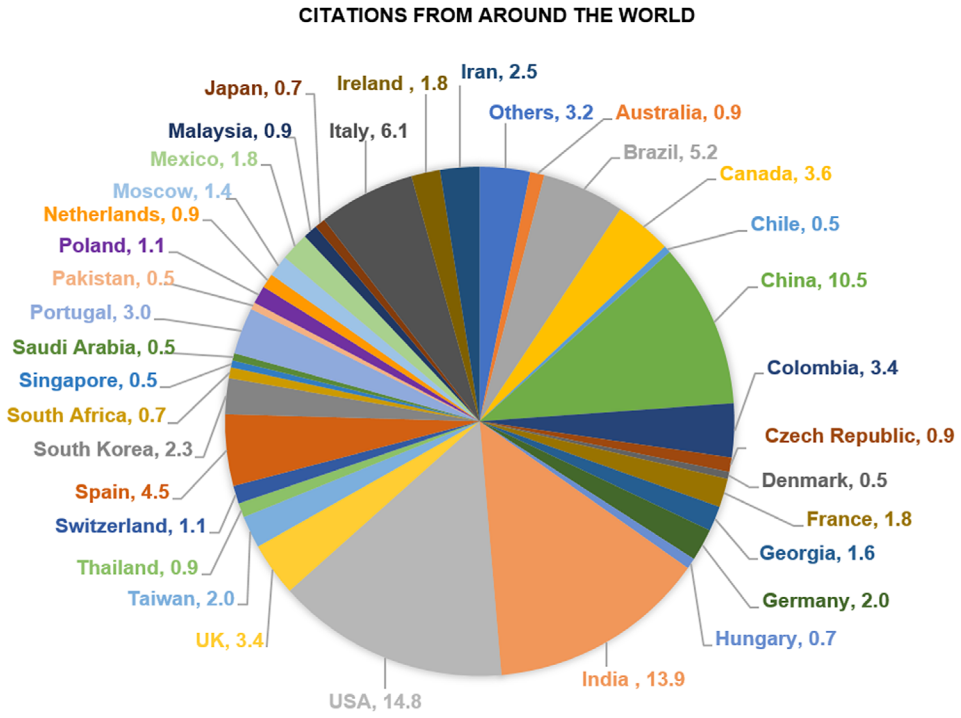

FIGURE 3 Country-wise share of citations (in %) of CAMP and its derivatives. Countries with fewer share of citations (<0.5%) were grouped as Others and these included Algeria, Argentina, Belgium, Cuba, Croatia, Ecuador, Finland, Egypt, Uruguay, Tunisia, Sweden, Russia, Norway, and Indonesia. CAMP, collection of antimicrobial peptides

5 | CONTRIBUTION OF CAMP, CAMPSIGN, AND CLASSAMP TO RESEARCH ON AMPs

5.1 | Creation of other AMP databases

The data present in CAMP was used to create other AMP databases like ADAM—a database of AMPs,[30] InverPep—a database of invertebrate AMPs,[31] YADAMP—yet another database of AMPs,[32] LAMP—a database linking AMPs,[33] C-PAmP—a database containing computationally predicted AMPs from plants,[34] Hemolytik—a database of experimentally determined hemolytic and nonhemolytic peptides,[35] dbAMP—a resource for exploring AMPs with functional activities and physicochemical properties on transcriptome and proteome data,[36] and ANTISTAPHYBASE—a database of AMPs and essential oils against methicillin-resistant *Staphylococcus aureus* (MRSA) and *Staphylococcus aureus*.[37]

5.2 | Creation of prediction algorithms on antimicrobial activity

The data present in CAMP have been used either for training or testing algorithms developed for AMP prediction and/or classification.[38–45]

5.3 | Identification of peptides from natural sources

The AMP prediction algorithm, available through CAMP, has been successfully used to identify/predict peptides with antimicrobial activity from natural sources such as *Protaetia brevitarsis* larvae,[46] Sichuan pepper,[47] *Litopenaeus vannamei*,[48] milk proteins,[49] human sweat,[50] *Thermophilic geobacillus* sp. Strain ZGt-1,[51] *Varanus komodoensis* (Komodo Dragon),[52] American alligator plasma,[53] human basal tear sample,[54] *Oxya chinensis sinuosa* (grasshopper),[55] lily leaves,[56] *Chrysochromulina tobin*,[57] and marine mussels.[58] Few of these peptides have been experimentally validated using wet-lab methods.

5.4 | Rational design of AMPs

Using CAMP data, Joker,[59] an algorithm was developed that aids in rational design of AMPs. CAMP with BLAST tool has been widely used to identify AMP sequences homologous to the user-defined sequences.[60–66] SVM model of CAMP was used for prediction of 10,000 double mutants of Bactenecin 2A and subsequently 17 peptides were shortlisted for experimental validation.[67]

5.5 | Relatedness of novel peptides to AMP families

The novelty of an antiviral peptide identified from the Asian medicinal plant *Acacia catechu* was evaluated by comparing it with members of the AMP families present in CAMPSign.[68] Differentially expressed peptides from serous ovarian cancer tissues were found to exhibit similarity to the members of the aurein AMP family based on sequence analysis using CAMPSign.[69]

6 | CONCLUSIONS

$CAMP_{R3}$, ClassAMP, and CAMPSign are available online at www.camp.bicnirrh.res.in, www.bicnirrh.res.in/classamp, and www.campsign.bicnirrh.res.in, respectively. Open access to CAMP and its derivatives has accelerated research on AMPs. These resources have improved AMP identification, prediction, and rational design. The citation reports are indicative of the world-wide usage of these resources (Figure 3). We hope to improve the functionality of these resources, as more data on AMPs are made available in the public domain.

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CONFLICT OF INTEREST

None declared.

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