The Karyotype Ontology: a computational representation for human cytogenetic patterns
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ABSTRACT
The karyotype ontology describes the human chromosome complement as determined cytogenetically, and is designed as an initial step toward the goal of replacing the current system which is based on semantically meaningful strings. This ontology uses a novel, semi-programmatic methodology based around the 

The ontology is available at http://www.purl.org/ontolink/karyotype/ The clojure code is available at http://code.google.com/p/karyotype-clj/

1 INTRODUCTION
A karyotype describes the chromosome complement of the individual. It can be easily assayed cytogenetically and, therefore, has been widely used as a mechanism for understanding the underlying genetic complement of cells and organisms. It remains of vital diagnostic importance, as well as a key tool for a large research community. Human karyotypes are normally represented using a string, as defined by the International System for human Cytogenetic Nomenclature 2009 (ISCN2009) (Shaffer et al., 2009) — here we call these ISCN strings. Unlike similar string-based representations such as InCHI (McNaught, 2006), ISCN strings lack a formal interpretation, and do not have good computational properties. For example, they cannot be represented in ASCII as they include meaningful underlining. Even the ISCN specification has no electronic representation and is not searchable.

In this paper, we describe our work in developing an ontological representation for karyotypes. Currently, karyotypes have only been represented as experimental entities, or results of medical procedures. The purpose of our ontology is to provide a strong computational and formal interpretation for a karyotype. This will enable semantic (and syntactic) checking of karyotypic information at the point of generation; it will allow the development of a knowledge base of karyotypes which is open to rich querying, and finally a web-capable interchange format as many different groups around the world generate this information.

2 WHAT IS AN ISCN STRING
ISCN defines a string format, initially designed for writing and printing, which provides a representation of the chromosome complement of a human, as determined cytogenetically by the banding patterns which are revealed after staining and fixation of metaphase cells. The ISCN specification has a long history. Initially, it was developed to address the need for an explicit nomenclature “to enable communication between workers in the field”. The early versions date from around 1960, when the emphasis was on human-to-human communication, and for small numbers of karyotypes.

ISCN strings represents a number of key concepts:

- The autosomal chromosomes are represented by a number 1 to 22.
- The sex chromosomes are represented by X or Y.
- Chromosomal structural components are represented: The long and short arm are represented by q and p; centromeres are represented by cen or more specifically, p10 for the part of the centromere facing the short arm or q10 for part facing the long arm; and telomeres are represented by ter.
- Bands at different resolutions are represented numerically such as 1p11.1.
- Changes from the base karyotypes are represented: del represents a deletion, add represents additional material.

In addition to these concepts, there are many more that can be represented in an extended karyotype: these include chromosomal groups, mosaicism, ploidy level and so forth. The full specification, describes many parts of human cytogenetics, including both the biology and the experimental techniques used. As would be expected for a specification with a long history, not all parts are regularly used.

Banding Patterns used to describe chromosome locations are defined cytogenetically by the appearance of the chromosome, during a part of division, following staining with a dye; this staining process is normally lethal to the cell. The original banding pattern described in the Paris Conference 1971 report, represented the results of three whole chromosome banding techniques: Quinacrine- (Q-), Giemsa- (G-), and Reverse-banding (R-).

The main components of stained chromosomes are:

- A band is a part of the chromosome that is distinguishable from its adjacent segments, appearing darker or lighter. Bands proximal to the centromere are labelled as 1, then 2 and so on.
- A region is an area of a chromosome lying between two landmarks. Regions adjacent to the centromere are labelled as 1 in each arm, then 2 and so on.
• A landmark is a consistent and distinct morphological feature of a chromosome. They are used as delimiters for regions.

Bands are represented numerically such that bands are numbered from the centromere outward. The band name is a combination of: the chromosome number, the arm symbol, the region number, and the band number within that region. For example, the band 1q42 is found on the long arm of chromosome 1 and is the second band, proximal to the centromere in region 4. Broadly speaking there is a correlation between the cytogenetic bands and the underlying DNA sequence of the chromosome; however, the very different scales (1q42 is 12.4Mb long) of these two measurements means that this relationship is approximate.

Cytogenetic banding also comes at several resolutions: high-resolution banding involves the staining of chromosomes during prophase, prometaphase, or interphase when the chromosomes are less condensed and spread over a large area; low-resolution uses the highly-condensed metaphase chromosome. The level of resolution is determined by the number of bands seen in a haploid set and ranges approximately from 300 to 850. High-resolution banding techniques result in existing bands being subdivided into sub-bands. Whenever a band is subdivided, a decimal place is placed after the original band number, and the sub-band number is appended to the band name with proximal sub-bands being labelled 1, then 2 and so on. For example, the sub-bands of band 1q42 will be: 1q42.1, 1q42.2, and 1q42.3, such that sub-band 1q42.1 is more proximal to the centromere. However when a sub-band is subdivided, then no additional decimal is added. For example, the sub-bands of sub-band 1q42.1 are: 1q42.11, 1q42.12, and 1q42.13.

Typical queries that we might wish to make of a collection of karyotypes include:

- Which karyotypes have abnormalities in a given chromosome?
- Which karyotypes increase the copy number of a given band?
- Which karyotypes affect a given band in any way?

Currently, these are hard to answer computationally because of the complexity of the ISCN strings, as well as intrinsic complexity of the biology. Our karyotype ontology aims to address the former, and contain the latter.

3 OUR METHODOLOGY

For the karyotype ontology, we have a very specific requirement which is to enable machine interpretation of the knowledge that is currently represented in ISCN strings. One of the implications of this is that our knowledge capture is extremely contained; virtually all the knowledge we require is present in the ISCN2009 specification. Our task is to formalise and represent this.

Our initial experiments with a realist ontology showed a number of difficulties; the distinction between a chromosome (as a piece of DNA and protein), the experimental artefact (following staining) and the visualisation of the experimental artefact are all different "portions of reality"; for instance, a chromosome in a live cell cannot meaningfully be said to have bands. These distinctions can be represented ontologically, however the result is a ontology with many duplicated hierarchies: chromosome 1, stained chromosome 1, and the visualisation of chromosome 1.

As these distinctions are not required for our application, we have instead followed a pragmatic approach (Lord and Stevens 2010). We have developed a lightweight ontology with specific computational goals. Our desire for computational support and inferencing, as well as a web-capable interchange format, has lead us to adopt OWL2 as our representation format.

While avoiding a realist approach has reduced some duplication, karyotype ontology still requires a considerable number of highly similar concepts, which is intrinsic to the problem domain. Trivially, for instance, the human karyotype requires 24 individual chromosome concepts, with similar logical and textual definitions. In turn, each chromosome has a complex band pattern (over 850 bands in total), including band intervals for use at different resolutions. Developing this type of ontology would be complex, time consuming and difficult to maintain using conventional tools. Therefore, we developed the tawny library, which allows fully programmatic development of OWL ontologies (Lord 2013). This library allows expansion of arbitrary patterns; this is similar to the capabilities of OPPL (Egana Aranguren et al. 2009), populous (Jupp et al. 2010) or safe macros (Mungall et al. 2010). However, it additionally provides us with the ability to define unit tests to provide computationally checkable expressions of the requirements for inferencing, a semi-literate programming environment, and the ability to make arbitrary syntactic extensions. The syntax is modelled after and highly similar to Manchester syntax, therefore it is presented here without further explanation; a fuller description is provided in the tawny documentation.

The basic structure of our ontology is shown in Figure 1.

![Fig. 1. The abstract structure of the karyotype ontology.](https://github.com/phillord/tawny-owl)
organism or cell line all (normal) Chromosome 21s have (visible) satellites, an individual chromosome, in an individual cell may not. Similarly small supernumerary marker chromosomes (sSMCs) cannot be fully represented in ISCN2009 (Liehr, 2009). This means that an ontological representation of the ISCN2009 specification cannot therefore be a completely faithful representation.

4 REPRESENTING ABNORMALITIES

Our initial experiments attempt to represent karyotypes as a rich partonomy, using the concepts described in the previous section. For example, the normal male karyotype 46,XY would be described as having 46 chromosomes of the appropriate types. One problem with this approach is that the definition of any karyotype is relatively large; while tawny enables the generation of this form of concept, it cannot reduce the complexity for reasoners; so this form of ontology is not likely to scale well. Further, a simple partonomy is not a rich enough representation, as it cannot represent simple inversions which contain all the same parts, but not necessarily in the right order. While it is possible to represent order in OWL (Drummond et al., 2006), this would add more complexity and scaling issues.

Further, there are some strong edge cases that are not complex, but impossible to represent as a partonomy; for example, the karyotype 45,X,-Y describes the chromosomes of a cell line, isolated from a normal male, which has lost its Y chromosome. This is a different karyotype but is partonomically indistinguishable from 45,X, a phenotypically female individual with Turner’s syndrome. Therefore we represented karyotypes using events: a karyotype is described as a set of changes from a base or normal karyotype. For example, 45,X,-Y is described as a 46,XY male, with a deletion event of Y, while 45,X female is described as a 46,XN, with a deletion event of a sex chromosome (see Listing1): in this case N represents an unknown autosome. OWL can represent partial knowledge straightforward.

Listing 1. A basic class definition

In total there are 13 events that represent the following key concepts:

- Addition: Chromosome gain or band addition.
- Deletion: Chromosome loss or band deletion, including terminal deletion with break (;) and interstitial deletion with breakage and reunion (:).
- Duplication: Band duplication. Specialised with DirectDuplication or InverseDuplication.
- Fission: Centric fission.
- Insertion: Band insertion between chromosomes. Specialised with DirectInsertion or InverseInsertion.
- Inversion: Band inversion, both paracentric and pericentric.
- Quadruplication: Band quadruplication.
- Translocation: Band translocation between chromosomes.
- Triplication: Band tripllication.

These concepts are supported by a number of properties created for the purpose, such as isBandOf.

The simplest representation in our ontology shares some limitations with the ISCN “short system” – a usable subset of ISCN, which is more generally used. For example, with both triplication or quadruplication events, we do not represent the orientation of all the repeats. It would be possible to differentiate these as two direct duplications, or one direct and one inverted duplication (for triplication); however, again it is useful to represent partial information; for many existing ISCN strings which use the short system, this knowledge is not available.

5 DEFINING SEX

One interesting outcome of both our representation and normal custom within cytology is that the definition of the sex of a karyotype is quite different from what might be expected. Intuitively, male would be defined as a karyotype with a Y chromosome[1] while female would be defined as a karyotype without. However, this intuitive definition is not correct. For example, the previously described 45,X,-Y has no chromosome Y and yet would generally be considered to be a male karyotype, since the organism from which the cell line originated was male. Our definitions of male and female, therefore, consider the “history” of the karyotype. Female is defined as derived from 46,XX (Listing2). Male from 46,XY (Listing1). This definition also copes with Turner’s syndrome which is not defined as either male or female, nor describe sex for haploid karyotypes: these can contain a Y chromosome (or not), but sex is not meaningful for these karyotypes. Karyotypes which are definitional for syndromes such as Turner’s or being male, are categorised under NamedKaryotype.

Listing 2. Definition of Female Karyotype

In total there are 13 events that represent the following key concepts:

3 As well as variation at a genetic level in even a clonal population, the cytogenetic definition of satellite is a staining region. So even for two genetically identical chromosomes, one may show a satellite, and another may not.

5 We ignore Y chromosome translocations for simplicity.
to determine the sex from the components of a karyotype; for future work, we may be able to address this, by describing the origin of the karyotype ($45$, $X$, $6$ is only valid as the karyotype of a cell line).

6 ASSESSMENT

As well as providing a specification, we are fortunate that ISCN2009 provides many examples; we are using these examples as an initial evaluation for our ontology, to determine whether the ontology is expressive enough to represent these exemplar karyotypes.

We wished this to be related to the ISCN string as, in most cases, there is no other more humanly readable name. In order to represent a karyotype in tawny a name is needed which is “safe” both as a URL and in Clojure, the language used to implement tawny, and, pragmatically, in Manchester syntax also.

- All karyotypes start with a “k” — Clojure symbols cannot start with numbers
- Replaced ; character with , — comment in Clojure
- Replaced ( and ) characters with ! — list delimiter in Clojure
- Replaced . character with , — separator in Manchester syntax

Currently, we have represented 71 karyotypes in our karyotype ontology. During this process, we have also discovered two difficulties with the existing ISCN2009 specification; in both cases a simple and intuitive correction is possible. These are:

- The lack of a band $Xq12$ in figures showing chromosome bands (page 31); the figures are the only list of all chromosome bands in ISCN2009.
- The absence of a band $Yq11.2$ in the 300 band resolution ($11.21, 11.22, 11.23$ do exist on page 31) while this band is used in several exemplars (for example on page 78). This band does exist in ISCN2005 – the previous specification.

Taken together, these 71 karyotypes use all of the distinctions necessary to answer questions given in Section 2.

7 DISCUSSION

The development of a karyotype ontology is potentially valuable for cytogenetics, as the current ISCN specification is not computationally amenable, reducing the value of collections of karyotypes as they are hard to query, check and maintain. The work described here presents an initial step towards this goal. The process of producing this ontology is already of use; we have discovered some errors or inconsistencies within ISCN which have found some errors or inconsistencies within ISCN which prevent its direct interpretation computationally; we expect to find more as we continue.

We have found the use of an ontology to be an appropriate mechanism; the knowledge that needs to be represented is complex, and overlapping. Cytogenetic data also requires the representation of partial knowledge, such as locations that are only known to a given resolution. The open world assumption of OWL copes well with this situation. Cytogenetics databases are also relatively small (100,000’s rather than millions or billions), sizes to which OWL should scale.

Existing tools for ontology development are, however, rather limited in their support for building this form of ontology; it is to address this need that we have developed tawny. This has proven to be highly useful: for example, the current karyotype ontology consists of 1466 classes, of which 1293 are used to represent the chromosomes and their bands at different resolutions. All of these classes have been generated from simpler data structures in Clojure. Additionally, the arbitrary expressiveness of tawny has allowed us to add syntax specific for the karyotype; many definitions in our ontology follow patterns. Using tawny these can be encoded as Clojure functions, such as that shown in and used in Listing 4.

```clojure
(defn inversion
  [n band1 band2]
  (exactly n hasEvent
   (owl: Inversion
     (owlsome hasBreakPoint
      band1 band2))))
```

Listing 4. A function used to define inverse events

In addition to the convenience, this also aids significantly in maintainability, as it is possible to change definitions for all classes that use this function. In time, we expect to extend this work to present an end-user syntax and parser, probably built directly using Clojure. Through the use of tawny we aim to build an end-to-end solution for the computational encoding of karyotypes.

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REFERENCES

Drummond, N., Rector, A., Stevens, R., Moulton, G., Horridge, M., Wang, H. H., and Seidenberg, J. (2006). Putting OWL in order: Patterns for sequences in OWL. Concrete, pages 1–10.

Egana Aranguren, M., Stevens, R., and Antezana, E. (2009). Transforming the axiomisation of ontologies: The ontology pre-processor language. Nature Precedings.

Jupp, S., Horridge, M., Iannone, L., Klein, J., Owen, S., Schanstra, J., Stevens, R., and Wolstencroft, K. (2010). Populos: A tool for populating ontology templates. Proceedings of the 3rd International Workshop on Semantic Web Applications and Tools for the Life Sciences BerlinGermany December 810 2010.

Licht, T. (2009). Small supernumerary marker chromosomes (sSMCs): a spotlight on some nomenclature problems. Journal of Histochemistry and Cytochemistry, 57(11), 991–993.

Lord, P. (2013). The Semantic Web takes Wing: Programming Ontologies with Tawny. OWL. http://arxiv.org/abs/1303.0213

Lord, P. and Stevens, R. (2010). Adding a little reality to building ontologies for biology. PLoS One.

McNaught, A. D. (2006). The IUPAC International Chemical Identifier (InChI). Chemistry International, 2006(September 18).

Mungall, C., Ruttenberg, A., and Osumi-Sutherland, D. (2010). Taking shortcuts with OWL using safe macros. Nature Precedings.

Shaffer, L., on Human Cytogenetic Nomenclature. J. S. C., Slovak, M., and Campbell, L. (2009). ISCN 2009: An International System for Human Cytogenetic Nomenclature (2009). Karger.