

WATSON-CRICK PAIRING, THE HEISENBERG GROUP AND MILNOR INVARIANTS

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ABSTRACT. We study the secondary structure of RNA determined by Watson-Crick pairing without pseudo-knots using Milnor invariants of links. We focus on the first non-trivial invariant, which we call the Heisenberg invariant. The Heisenberg invariant, which is an integer, can be interpreted in terms of the Heisenberg group as well as in terms of lattice paths.

We show that the Heisenberg invariant gives a lower bound on the number of unpaired bases in an RNA secondary structure. We also show that the Heisenberg invariant can predict *allosteric structures* for RNA. Namely, if the Heisenberg invariant is large, then there are widely separated local maxima (i.e., allosteric structures) for the number of Watson-Crick pairs found.

1. INTRODUCTION

Ribonucleic acid (RNA) is a nucleic acid polymer consisting of nucleotide monomers, each of which is of one of four types determined by the nucleotide base present in it. RNA plays a central role in living cells, specifically in the synthesis of proteins using DNA as a template. In addition, RNA itself can serve as an information carrier and also has catalytic properties. Indeed its ability to serve as both an information carrier and a catalyst has led to speculation that life began as an *RNA world* [12].

The *primary structure* of an RNA molecule is the sequence of nucleotide bases in it. In addition to this, the properties of an RNA molecule depend strongly on the *secondary structure*, which is the 3-dimensional shape of the molecule [13].

The bases form two complementary pairs, with members of a pair forming strong hydrogen bonds. Thus, if two subsequences of the RNA sequence are complementary, a stable secondary structure called a *stem loop structure* can be formed by bonds between complementary pairs in these subsequences. We study here these *secondary structures* of an RNA molecule, determined by the Watson-Crick pairing. We shall consider structures without so called *pseudo-knots*. For basic concepts for RNA folding, we refer to [27] and [12]. For surveys and other studies of RNA folding, we refer to [12]–[28]. Henceforth by secondary structure we mean the secondary structure determined by Watson-Crick pairing without pseudo-knots.

We focus here on introducing new methods to yield a conceptual understanding of RNA folding. The model we consider - Watson-Crick pairing without pseudo-knots, is clearly an approximation in various ways. Firstly, stereo-chemical forces do not allow very short loops. Secondly, pseudo-knots are present in nature. Thus, allowing short loops but not pseudo-knots is an approximation of stereo-chemical

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forces. Further, we do not take into account the difference between the strengths of the A-U and the G-C bonds, and also ignore the non-Watson-Crick bonds.

Thus, our model is clearly not appropriate for the computational study of individual RNA molecules. Our goal is rather to introduce new methods for understanding RNA secondary structure, and show that these are very powerful in the context of our model. One can extend these methods to take into account more realistic models of RNA secondary structure.

Our methods can be motivated by a simple observation – if, for example, there are more Adenine than Uracil bases, then some Adenine bases must be unpaired. These considerations give a very elementary lower bound on the number of unpaired bases. Mathematically, this can be viewed as coming from abelianisation. Another context in which abelianisation gives the simplest criteria is the *linking number* for classical links. In this paper, we show that one can adapt Milnor’s theory of *higher linking numbers* to the context of RNA.

We use a natural model for Watson-Crick pairing of RNA (without pseudo-knots) in terms of the free group F on two generators α and β . An element of the free group is given by a word in the four letters α , $\bar{\alpha}$, β and $\bar{\beta}$, with $\bar{\alpha}$ and $\bar{\beta}$ the inverses of α and β respectively. We identify these letters with the nucleotides Adenine, Uracil, Guanine and Cytosine respectively. Under this identification, an RNA molecule gives a string in the four letters α , β , $\bar{\alpha}$ and $\bar{\beta}$.

Stem loops, which are the basic units of RNA secondary structure, then correspond to words in the free group of the form $gl\bar{g}$, with g and l words in the free group and \bar{g} the inverse of g (see figure 1). One may further have Watson-Crick pairing within the word l , so that a subword of the RNA sequence may be of the form $ga(hl\bar{h})b\bar{g}$ as in figure 2. In general, a secondary structure without pseudo-knots consists of pairings between letters and their inverses so that there is no *nesting*. We formalise such a structure (which we call a *folding*) in Definition 5.1. The appropriate *energy*, whose local and global minima we study, is the number of unpaired bases in the secondary structure.

Mathematically, one can interpret the above as saying that the number of unpaired bases is a *conjugacy invariant norm*. For, a secondary structure on RNA corresponding to the word w gives one for the word $gw\bar{g}$ (with the initial segment corresponding to g paired with the final segment corresponding to \bar{g}) so that the number of unpaired bases is the same. Further, given secondary structures on strands of RNA corresponding to words w_1 and w_2 , we get a secondary structure on the strand corresponding to w_1w_2 (the concatenation of the words). This paper is based on the observation that conjugacy invariance can be taken into account by considering nilpotent quotients.

In analogy with Milnor invariants, we shall associate numbers to words g in the letters α , β , $\bar{\alpha}$ and $\bar{\beta}$, which we call *invariants* according to terminology familiar in mathematics (they are not to be taken as invariant in any biological sense). By the above, it is desirable that they are invariant under conjugation, or at least the extent to which they are not invariant can be estimated. To achieve sub-additivity, we consider additive functions of g and take their absolute value, or more generally sums of the absolute values of such functions.

As we have seen, an obvious lower bound for the number of unpaired elements is given by comparing the number of letters that are α with the number that are $\bar{\alpha}$ and similarly for β and $\bar{\beta}$. We denote the difference between the number of

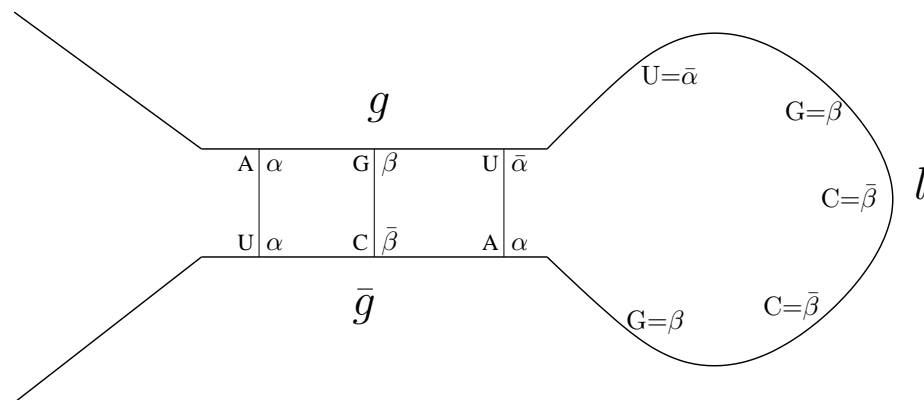


FIGURE 1. A stem loop with bases labelled using both nucleotides and letters in the free group

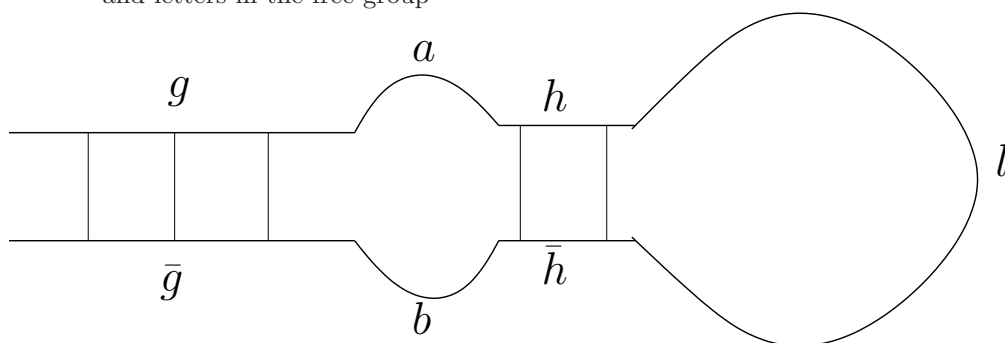


FIGURE 2. An RNA secondary structure

letters of a word g that are α and the number that are $\bar{\alpha}$ by $a(g)$. Similarly, $b(g)$ denotes the difference between the number of letters that are β and the number that are $\bar{\beta}$. In other words we look at the image of g under the abelianisation map $ab : F \rightarrow \mathbb{Z}^2$. Then $a(g)$ and $b(g)$ denote the co-ordinates of $ab(g)$ so that $ab(g) = a(g)ab(\alpha) + b(g)ab(\beta)$. The numbers $a(g)$ and $b(g)$ are the first of our invariants.

Clearly, the minimum number of unpaired bases in g is at least $|a(g)| + |b(g)|$, but this is very far from sharp. We proceed further in analogy with Milnor's theory of link homotopy. The simplest invariants of a link are the linking numbers. These are given by considering the image of a curve in the abelianisation of an appropriate fundamental group, and are thus analogous to $a(g)$ and $b(g)$. Milnor constructed *higher linking numbers* by considering appropriate nilpotent quotients.

We shall associate an appropriate link to RNA molecules in Section 10 and construct various invariants. The main focus in this paper, however, is to construct and study the first of these higher invariants. It is easiest to proceed with a direct algebraic description. This description is in terms of another familiar object – the Heisenberg group. We thus call this invariant the *Heisenberg invariant* $\nu(g)$. There is also a nice geometric view of this invariant in terms of areas enclosed by *lattice*

paths. We also provide an elementary combinatorial description in Theorem 6.1 which allows for easy computation.

We show that the Heisenberg invariant gives a lower bound on the number of unpaired bases in an RNA molecule, and hence the potential energy of a secondary structure.

We also show that the Heisenberg invariant is related to a biologically significant property of Watson-Crick pairing. Namely, we show that if $\nu(g)$ is *sufficiently large*, then there are *allosteric structures*, i.e., local minima for the number of unpaired bases that are *widely separated*. This means that there are two ways of *folding* the sequence so that we cannot pass from one to the other without significantly increasing the number of unpaired bases (here a folding is an abstraction of the secondary structure). Thus, the Heisenberg invariant as well as the higher invariants should prove very fruitful in the study of RNA secondary structures.

Our results depend on our simplified model. However, it is easy to see that similar results continue to hold even if one makes the model biologically more realistic by taking into account that nearby bases do not pair (i.e., there are no very short loops). In the case of the lower bound this is obvious, as further restrictions can only increase the number of unpaired bases. The result concerning allosteric structures also extends as we sketch following the proof of the result for our model.

2. THE HEISENBERG INVARIANT

The Milnor higher link invariants [9][10] are based on the lower central series of a group. We recall some basic definitions.

Consider a group G . For elements $a, b \in G$, \bar{a} denotes the inverse of a and $[a, b]$ denotes the commutator $ab\bar{a}\bar{b}$. For subgroups $H_1, H_2 \subset G$, we define $[H_1, H_2]$ to be the normal subgroup generated by elements of the form $[a, b]$, $a \in H_1, b \in H_2$.

The lower central series of G is defined inductively as follows. Let $G_1 = G$. If G_n has been defined, we define G_{n+1} to be $[G, G_n]$. In particular, $G_2 = [G, G]$ and the abelianisation of G is G/G_2 . Note that $G_1 \supset G_2 \supset G_3 \supset \dots$

Consider now the free group F generated by α and β . Let H be the group F/F_3 . The Heisenberg invariant is obtained by considering the image $[g] \in H$ of an element $g \in F$. It is well known that this is the Heisenberg group, which is the unique central extension of \mathbb{Z}^2 by \mathbb{Z} . For the convenience of the reader, we prove these properties below.

Proposition 2.1. *There is an exact sequence*

$$(1) \quad 1 \rightarrow \mathbb{Z} \rightarrow H \rightarrow \mathbb{Z}^2 \rightarrow 1$$

with the image of \mathbb{Z} central in H and generated by $[\alpha, \beta]$. Further, H is isomorphic to the Heisenberg group.

Proof. As $F_3 \subset F_2$, the abelianisation homomorphism $ab : F \rightarrow \mathbb{Z}^2 = F/F_2$ induces a surjective homomorphism $\varphi : H = F/F_3 \rightarrow F/F_2 = \mathbb{Z}^2$. It is well known that the kernel of $ab : F \rightarrow \mathbb{Z}^2$ is the normal subgroup in F generated by $[\alpha, \beta]$. Hence the kernel of φ is the normal subgroup in H generated by the equivalence class of $[\alpha, \beta]$ in H , which we continue to denote by $[\alpha, \beta]$.

Note that the commutators $[[\alpha, \beta], \alpha]$ and $[[\alpha, \beta], \beta]$ are both elements of F_3 , and hence have trivial images in H . It follows that $[\alpha, \beta] \in H$ commutes with the images of α and β in H and hence is central. Thus, as the kernel of φ is the normal subgroup generated by φ , it is in fact the cyclic group generated by $[\alpha, \beta]$.

To complete the proof of the exact sequence of Equation 1, it suffices to show that no power of $[\alpha, \beta]$ is trivial in the group H .

We show this by constructing an explicit homomorphism from H to the Heisenberg group \mathbb{H} with integer coefficients, namely the group of matrices of the form

$$(2) \quad M(a, b, c) = \begin{pmatrix} 1 & a & c \\ 0 & 1 & b \\ 0 & 0 & 1 \end{pmatrix}$$

with a, b and c integers.

The homomorphism $\psi : H \rightarrow \mathbb{H}$ is defined as follows. Let $\Psi : F \rightarrow \mathbb{H}$ be the unique homomorphism taking α and β to the matrices $M(1, 0, 0)$ and $M(0, 1, 0)$. By a well known (and straightforward) computations, $\Psi([\alpha, \beta]) = M(0, 0, 1)$, and $M(0, 0, 1)$ is central in \mathbb{H} . Hence $\Psi(F_3)$ is the trivial group. Thus, we get a well-defined homomorphism $\psi : H = F/F_3 \rightarrow \mathbb{H}$, which is clearly surjective. As $\psi([\alpha, \beta]) = M(0, 0, 1)$, it follows that $\psi([\alpha, \beta]^k) = M(0, 0, k)$. Hence if $k \neq 0$, $[\alpha, \beta]^k$ is non-trivial as an element of H . This shows that the sequence of Equation 1 is exact.

Finally, we show that ψ is injective, hence an isomorphism. Suppose $\psi(g) = 1$. Then as ψ gives an isomorphism on the abelianisations of H and \mathbb{H} , it follows that $ab(g) = 0$. Hence by the exact sequence, $g = [a, b]^k$ for some $k \in \mathbb{Z}$. It follows that $\psi(g) = M(0, 0, k)$, hence $\psi(g) = 1 \implies k = 0 \implies g = 1$. \square

The homomorphisms $a : F \rightarrow \mathbb{Z}$ and $b : F \rightarrow \mathbb{Z}$ defined on F factor through H (as $F_3 \subset F_2$), and we continue to denote them by $a(\cdot)$ and $b(\cdot)$. We shall also denote the images of α and β in H by α and β .

It is easy to see that for an element $g \in H$, $a(g)$ and $b(g)$ are the entries a and b of $\psi(g)$. We can define the Heisenberg invariant in terms of the remaining entry c . However, it will be convenient to take a different approach based on the following proposition (which is a special case of normal forms that are well known in the literature).

Proposition 2.2. *Any element $g \in H$ can be uniquely expressed as*

$$g = [\alpha, \beta]^\nu \alpha^a \beta^b$$

with ν, a and b integers.

Proof. Let $h = g\beta^{-b(g)}\alpha^{-a(g)}$. Then the image under the abelianisation map $\varphi(h)$ of h is trivial. Hence h is in the kernel of φ . By Proposition 2.1, $h = [a, b]^\nu$ for a unique ν . Hence it follows that $g = [\alpha, \beta]^\nu \alpha^a \beta^b$ with $a = a(g)$ and $b = b(g)$.

To see uniqueness, observe by abelianising that if $g = [\alpha, \beta]^\nu \alpha^a \beta^b$, we must have $a = a(g)$ and $b = b(g)$. Further, if $h = g\beta^{-b}\alpha^{-a}$, then ν is the unique integer such that $h = [a, b]^\nu$, and hence is determined by g . \square

Definition 2.3. The *Heisenberg invariant* of $g \in H$ is the unique ν such that $g = [\alpha, \beta]^\nu \alpha^a \beta^b$ with a and b integers. The Heisenberg invariant of a word in F is the Heisenberg invariant of the image of the word in H .

In terms of the above definition, the representation of Proposition 2.2 can be expressed as

$$(3) \quad g = \alpha^{a(g)} \beta^{b(g)} [\alpha, \beta]^{\nu(g)}$$

The higher link invariants are defined only for links with trivial linking number. In our situation, we have defined the Heisenberg invariant for all words. Nevertheless, it should be regarded as well-defined up to an error given by the abelianisation $(a(g), b(g))$. For instance, in Proposition 2.2, we can interchange the order of β and α in the representation of g . An analogous result still holds but we get a different value of the Heisenberg invariant.

We have the following simple properties of the Heisenberg invariant. In what follows all equalities are to be understood to be in the group H .

Proposition 2.4. *If $g_1, g_2 \in H$ have trivial abelianisations, $\nu(g_1g_2) = \nu(g_1)\nu(g_2)$.*

Proof. As g_1 and g_2 have trivial abelianisation, by Proposition 2.1 $g_i = [\alpha, \beta]^{k_i}$, $i = 1, 2$, for some integers k_i . By definition, $\nu(g_i) = k_i$. Further, $g_1g_2 = [\alpha, \beta]^{k_1+k_2}$, hence $\nu(g_1g_2) = k_1 + k_2 = \nu(g_1) + \nu(g_2)$. \square

Proposition 2.5. *If $g \in H$ has trivial abelianisation, then for $h \in H$, $\nu(hg\bar{h}) = \nu(g)$.*

Proof. By Proposition 2.1, as g is in the kernel of the abelianisation homomorphism φ , g is central. Thus, $hg\bar{h} = g$ \square

For the sake of clarity, we shall focus on elements $g \in H$ with trivial abelianisation. However, it is easy to obtain variants of all our results allowing for errors determined by $a(g)$ and $b(g)$ using the following proposition.

Proposition 2.6. *For $k \in \mathbb{Z}$, the following identities hold.*

- (1) $\nu(\alpha^k g) = \nu(g)$
- (2) $\nu(g\beta^k) = \nu(g)$

Proof. As $g = [\alpha, \beta]^{\nu(g)} \alpha^{a(g)} \beta^{b(g)}$ and $[\alpha, \beta]$ is central, $\alpha^k g = [\alpha, \beta]^{\nu(g)} \alpha^{k+a(g)} \beta^{b(g)}$, hence $\nu(\alpha^k g) = \nu(g)$. Similarly, $g\beta^k = [\alpha, \beta]^{\nu(g)} \alpha^{a(g)} \beta^{b(g)+k}$, hence $\nu(g\beta^k) = \nu(g)$. \square

We shall say that the word g is *balanced* if $a(g) = b(g) = 0$.

3. LATTICE PATHS AND AREA

In this section, we give a geometric interpretation of the Heisenberg invariant, and some obvious extensions that follow from this. Examples viewed in this fashion form the intuition for the rest of the paper. As the results here are not used formally elsewhere, we shall skip most proofs.

The plane \mathbb{R}^2 contains the lattice \mathbb{Z}^2 . We can associate to each word $g = l_1 l_2 \dots l_n$ in the letters $\alpha, \bar{\alpha}, \beta$ and $\bar{\beta}$ a path in the plane as follows. We start at the origin $(0, 0)$. In the first step, we take a path from $(0, 0)$ to one of the points $(1, 0)$, $(0, 1)$, $(-1, 0)$ and $(0, -1)$ according as l_1 is $\alpha, \beta, \bar{\alpha}$ or $\bar{\beta}$. Inductively, at the end of the $(k-1)$ th step we will have a path from $(0, 0)$ to a lattice point $(p, q) \in \mathbb{Z}^2$. We extend the path by a unit segment joining (p, q) to one of the points $(p, q) + (1, 0)$, $(p, q) + (0, 1)$, $(p, q) + (-1, 0)$ and $(p, q) + (0, -1)$ according as l_k is $\alpha, \beta, \bar{\alpha}$ or $\bar{\beta}$.

Thus, we obtain a path in the plane consisting of horizontal and vertical segments. If $a(g) = b(g) = 0$, this path is a loop γ . The Heisenberg invariant is the *oriented area*, interpreted appropriately, bounded by the loops γ . If γ is a simple loop, it bounds a region R . In this case, $\nu(g)$ is $\pm \text{Area}(R)$, with the sign determined by whether γ is a counterclockwise or a clockwise loop around R . In general,

we can regard γ as the boundary of a region R , allowing signs and multiplicities, which we define below in terms of winding numbers. The area of R , taking into account the signs and multiplicities, is the Heisenberg invariant $\nu(g)$.

Consider a unit square $\Delta(p, q)$, $p, q \in \mathbb{Z}^2$, with vertices (p, q) , $(p+1, q)$, $(p, q+1)$ and $(p+1, q+1)$. Let $c(p, q)$ be the winding number of γ about the centre z of $\Delta(p, q)$. We remark that we can take the winding number about any interior point to get the same result. The integer $c(p, q)$ is the (possibly negative) multiplicity of the square $\Delta(p, q)$.

Note that only finitely many of these coefficients are non-zero. We can interpret the Heisenberg invariant as (the finite sum)

$$\nu(g) = \sum_{(p,q) \in \mathbb{Z}^2} c(p, q)$$

In these terms, there are obvious extensions of the Heisenberg invariant. The group \mathbb{Z}^2 acts on the plane by translations. We use multiplicative notation for \mathbb{Z}^2 and denote generators by s and t , so that the action of s is translation by $(1, 0)$ and that of t is translation by $(0, 1)$. Let $\Delta = \Delta(0, 0)$. Then $\Delta(p, q)$ is the image $s^p t^q \Delta$ of Δ . The region R can be expressed as the formal sum

$$\sum_{(p,q) \in \mathbb{Z}^2} c(p, q) s^p t^q \Delta$$

Thus, we associate to g the polynomial in two variables

$$P_g(s, t) = \sum_{(p,q) \in \mathbb{Z}^2} c(p, q) s^p t^q$$

The Heisenberg invariant is $P_g(1, 1)$. Extensions are given by other (linear) functions of the polynomial P_g . As mentioned in the introduction, it is natural to consider such functions $F(P_g)$ so that if g is conjugate to g' , then $F(P_g) = F(P_{g'})$. This translates to being invariant under multiplication by the polynomial $s^k t^l$ for $k, l \in \mathbb{Z}$, i.e., for a polynomial P , $F(P) = F(s^k t^l P)$. It is clear that the Heisenberg invariant $P_g(1, 1)$ has this property. We next show how to construct secondary invariants, i.e., which are invariant under conjugation provided the Heisenberg invariant vanishes.

Let $p_g(s) = P_g(s, 1)$. Then $p_g(s)$ is invariant under multiplication by any power of t . Let $\nu_s(g) = p'_g(1)$. Suppose $\nu(g)$ vanishes. Then for an element g' conjugate to g , $P_{g'}(s, t)$ is of the form $s^k t^l P_g(s, t)$. Hence,

$$p'_{g'}(1) = (s^k p_g)'(1) = (k s^{k-1} p_g)(1) + (s^k p'_g)(1) = p'_g(1)$$

as $p_g(1) = \nu(g) = 0$ by hypothesis.

We have a similar invariant taking t in place of s . In case these invariants vanish, we get further invariants by taking higher derivatives.

We next turn to the general case, where we do not necessarily have $a(g) = b(g) = 0$. As before, we get a path from $(0, 0)$ to $(a(g), b(g))$. We make this into a loop γ by extending this by the vertical segment to $(a(g), 0)$ and then the horizontal segment to the origin. The Heisenberg invariant is then the area enclosed by this loop. Note that there are other minimal paths joining $(a(g), b(g))$ to the origin, which give different values for the Heisenberg invariant. Hence $\nu(g)$ should be regarded as defined up to indeterminacy given by $a(g)$ and $b(g)$.

We give another interpretation of the above in homological terms. This will not be used in the sequel.

The plane \mathbb{R}^2 has a natural structure as a cell complex X with vertices lattice points, edges horizontal or vertical unit segments joining adjacent lattice points and faces unit squares. The group \mathbb{Z}^2 acts on this cell complex freely by translations. The quotient Y of the one-skeleton $X^{(1)}$ under this action is the wedge of two circles. This has fundamental group F , and the one-skeleton of X is the (Galois) cover corresponding to the subgroup $F_2 = [F, F]$.

Any word g in the free group gives a path in Y . This lifts to a path in γ starting at the origin which can be regarded as a 1-chain in $C_1(X)$. If $a(g) = b(g) = 0$, then γ is a loop as $g \in F_2$, and hence is a 1-cycle. As the plane is contractible, this is a boundary $\gamma = \partial\zeta$, $\zeta \in C_2(X)$. As $H_2(X)$ and $C_3(X)$ are trivial, it follows that ζ is unique.

Let Δ be a fixed unit square. Any other unit square is the image $g\Delta$ of Δ under the action of \mathbb{Z}^2 on $C_2(X)$, and g is unique. As the unit squares are a basis of $C_2(X)$, we can uniquely express ζ as a finite sum.

$$\zeta = \sum_i n_i g_i \Delta, \quad n_i \in \mathbb{Z}, g_i \in \mathbb{Z}^2$$

We can interpret the Heisenberg invariant as

$$\nu(g) = \sum_i n_i$$

This has obvious extensions. We note that we can associate to g the element $\sum_i n_i g_i$ in the group ring $\mathbb{Z}[\mathbb{Z}^2]$ (which corresponds to the polynomial $P_g(s, t)$). This is well-defined (but is natural only up to multiplication by an element of the group \mathbb{Z}^2). We have considered the image of this element under the homomorphism $\mathbb{Z}[\mathbb{Z}^2] \rightarrow \mathbb{Z}$ taking each element of the group \mathbb{Z}^2 to 1. We can obviously obtain more refined estimate by considering either the full group ring, or at least other representation of the group ring.

4. IDENTITIES FOR THE HEISENBERG INVARIANT

We collect in this section some elementary identities in the group H and formulae for the Heisenberg invariant. Recall that the element $[\alpha, \beta]$ is central in the group H .

Lemma 4.1. *The following identities hold in H .*

- (1) $\alpha\beta\bar{\alpha} = [\alpha, \beta]\beta$.
- (2) For $k \in \mathbb{Z}$, $\alpha\beta^k\bar{\alpha} = [\alpha, \beta]^k\beta^k$.
- (3) $\beta\alpha\bar{\beta} = [\alpha, \beta]^{-1}\alpha$.
- (4) For $k \in \mathbb{Z}$, $\beta\alpha^k\bar{\beta} = [\alpha, \beta]^{-k}\alpha^k$.
- (5) For $g \in H$, $\alpha g\bar{\alpha} = [\alpha, \beta]^{b(g)}g$.
- (6) For $g \in H$, $\beta g\bar{\beta} = [\alpha, \beta]^{-a(g)}g$.

Proof. As $[\alpha, \beta]\beta = \alpha\beta\bar{\alpha}\bar{\beta}\beta = \alpha\beta\bar{\alpha}$, the first identity follows. As $[\alpha, \beta]$ is central, the second follows by taking a power.

Further, as $[\alpha, \beta]^{-1} = \beta\alpha\bar{\beta}\bar{\alpha}$, we have $\beta\alpha\bar{\beta} = [\alpha, \beta]^{-1}\alpha$. Once more we take a power to get the next identity.

Next, if $g \in H$, by Equation 3, $g = [\alpha, \beta]^{\nu(g)}\alpha^{a(g)}\beta^{b(g)}$, hence we can express its conjugate by α as $\alpha g\bar{\alpha} = (\alpha[\alpha, \beta]^{\nu(g)}\bar{\alpha})(\alpha\alpha^{a(g)}\bar{\alpha})(\alpha\beta^{b(g)}\bar{\alpha})$. As α commutes

with $[\alpha, \beta]$ and with powers of α , using the previous identities and that $[\alpha, \beta]$ is central, $\alpha g \bar{\alpha} = [\alpha, \beta]^{\eta(g)} \alpha^{a(g)} [\alpha, \beta]^{b(g)} \beta^{b(g)} = [\alpha, \beta]^{b(g)} g$ as claimed. The proof of the remaining identity is similar. \square

We deduce the effect of *canceling* a pair of letters in g that are not adjacent on the Heisenberg invariant.

Lemma 4.2. *Let g_1 and g_2 be words in H .*

- (1) $\nu(g_1 \alpha g_2 \bar{\alpha}) = b(g_2) + \nu(g_1 g_2)$
- (2) $\nu(g_1 \beta g_2 \bar{\beta}) = -a(g_2) + \nu(g_1 g_2)$

Proof. By Lemma 4.1, as $[\alpha, \beta]$ is central, $g_1 \alpha g_2 \bar{\alpha} = g_1 [\alpha, \beta]^{a(g_2)} g_2 = [\alpha, \beta]^{b(g_2)} g_1 g_2$. From this, it follows that $\nu(g_1 \alpha g_2 \bar{\alpha}) = b(g_2) \nu(g_1 g_2)$. The other identity is similar. \square

5. FOLDINGS AND THE HEISENBERG INVARIANT

Consider henceforth a fixed word $g = l_1 l_2 \dots l_n$ of length n in $\alpha, \bar{\alpha}, \beta$ and $\bar{\beta}$. We shall regard the letters as cyclically ordered, so that $l_{n+1} = l_1$. The word represents an RNA strand. There is an obvious description of RNA secondary structures without pseudo-knots in these terms.

Definition 5.1. A *folding* (or *fold*) of the word g is a collection of *disjoint* pairs $\mathcal{F} \subset \{(i, j) : 1 \leq i, j \leq n, i \neq j\}$ such that

- (1) For $(i, j) \in \mathcal{F}$, either $l_i = \alpha$ and $l_j = \bar{\alpha}$ or $l_i = \beta$ and $l_j = \bar{\beta}$.
- (2) For pairs $(i_1, j_1) \in \mathcal{F}$ and $(i_2, j_2) \in \mathcal{F}$, $i_1 < i_2 < j_1 \implies i_1 < j_2 < j_1$ and $i_1 > i_2 > j_1 \implies i_1 > j_2 > j_1$.

We denote the number of pairs in \mathcal{F} by $|\mathcal{F}|$. The folding process for RNA is governed by an attempt to maximise $|\mathcal{F}|$. We define a pair $(i, j) \in \mathcal{F}$ to be an α -pair if $l_i = \alpha$ and a β -pair if $l_i = \beta$. Every pair is either an α -pair or a β -pair.

The condition on the pairs rules out *nesting* (i.e., pseudo-knots). For a pair $(i, j) \in \mathcal{F}$, we define the word $w(i, j)$ between the letters as follows. If $i < j$, then $w(i, j) = l_{i+1} l_{i+2} \dots l_{j-1}$. If $i > j$, then $w(i, j) = l_{i+1} l_{i+2} \dots l_n l_1 l_2 \dots l_{j-1}$. Thus, this is the word from the i th letter to the j th letter in the counterclockwise direction in the cyclic ordering. The word $D_{ij}(g)$ obtained from g by *canceling* the pair $(i, j) \in \mathcal{F}$ is the word with $(n - 2)$ letters obtained by deleting l_i and l_j .

Using the conjugacy invariance of ν , we can rephrase Lemma 4.2 in the following way.

Lemma 5.2. *For $(i, j) \in \mathcal{F}$, if $l_i = \alpha$, then $\nu(D_{ij}(g)) = \nu(g) + b(w(i, j))$. If $l_i = \beta$, then $\nu(D_{ij}(g)) = \nu(g) - a(w(i, j))$*

Given a folding $\mathcal{F} = \{(i_1, j_1), (i_2, j_2), \dots, (i_k, j_k)\}$, we can inductively compute $\nu(g)$ as follows. Let $g_0 = g$ and let $g_1 = D_{i_1 j_1}(g)$. Note that \mathcal{F} induces a folding \mathcal{F}_1 of g_1 consisting of pairs of letters in \mathcal{F} other than (i_1, j_1) . We can thus continue inductively, deleting the pair corresponding to (i_2, j_2) . Thus, we get a sequence of words g_0, g_1, \dots, g_k .

Let $c(i, j) = b(w(i, j))$ if $l_i = \alpha$ and $c(i, j) = -a(w(i, j))$ if $l_i = \beta$. By inductively using Lemma 5.2, we get the following formula.

Lemma 5.3. *We have*

$$\nu(g) = \nu(g_k) + \sum_{l=1}^k c(i_l, j_l)$$

Proof. First note that by Lemma 5.2, $\nu(g) = \nu(g_1) + c(i_1, j_1)$. We shall iterate this process. To do so, observe that to apply Lemma 5.2 to g_1 , we need to consider the word $w_1(i_2, j_2)$ in g_1 between i_2 and j_2 . By the no nesting condition, this either equals $w(i_2, j_2)$ or differs from this by a pair of letters which are inverses of each other. In either case, $a(w_1(i, j)) = a(w(i, j))$ and $b(w_1(i, j)) = b(w(i, j))$. Thus, by Lemma 5.2, $\nu(g_1) = \nu(g_2) + c(i_2, j_2)$. We can now proceed inductively in this fashion to prove the claim. \square

An α -folding is a folding such that for $(i, j) \in \mathcal{F}$, $l_i = \alpha$. A β -folding is defined similarly. Consider a word g such that $a(g) = 0$. A *complete* α -folding is an α -folding \mathcal{F} such that if $l_i = \alpha$, then for some $j \neq i$, $(i, j) \in \mathcal{F}$. The following proposition is straightforward.

Proposition 5.4. *If $a(g) = 0$, there is a complete α -fold for g .*

Proof. If no letter is α (hence no letter is $\bar{\alpha}$ as $a(g) = 0$), we take \mathcal{F} to be the empty set. Otherwise we can find a pair (i, j) such that $w(i, j)$ does not contain the letters α and $\bar{\alpha}$. We can proceed inductively by considering the word $g' = w(j, i)$. This continues to satisfy the hypothesis and has fewer letters that are α than in g . A complete α -fold for g' together with (i, j) gives a complete α -fold for g . \square

We can analogously define complete β -folds. If $b(g) = 0$ there are complete β -folds. Observe that for a complete α -fold \mathcal{F} , $|\mathcal{F}|$ is the number n_α of letters that are α . Let n_β similarly denote the number of letters that are β .

Lemma 5.3 takes a particularly simple form for complete α -folds (and β -folds).

Lemma 5.5. *let g be a word with $a(g) = b(g) = 0$ and let \mathcal{F} be a complete α -fold for g . Then using the notation of Lemma 5.3*

$$\nu(g) = \sum_{l=1}^k b(w(i_l, j_l))$$

Proof. As \mathcal{F} is a complete α -folding, the word g_k consists of the letters of g that are β or $\bar{\beta}$. Thus, $g_k = \beta^{b(g)} = 1$ as $b(g) = 0$, hence $\nu(g_k) = 0$. As $l_i = \alpha$ for all pairs $(i, j) \in \mathcal{F}$, $c(i, j) = b(w(i, j))$. Thus the claim follows from Lemma 5.3 \square

6. COMPUTING THE HEISENBERG INVARIANT

The results of the previous section give an elementary formula for the Heisenberg invariant from which it can be readily computed. We formulate this below.

Let g be a word in the free group F . Let $w(0, i)$ denote the sub-word consisting of the $i - 1$ letters preceding l_i .

Theorem 6.1. *The Heisenberg invariant is given by*

$$\sum_{l_i=\bar{\alpha}} b(w(0, i)) - \sum_{l_j=\alpha} b(w(0, j))$$

Proof. The proof consists of inductively applying Lemma 5.2 and Proposition 2.6. Consider the letters $l_{i_1}, l_{i_2}, \dots, l_{i_k}$ of g that are either α or $\bar{\alpha}$. Let $g_0 = g$ and let g_1, g_2, \dots, g_k be obtained from g by successively deleting the letters $l_{i_1}, l_{i_2}, \dots, l_{i_k}$. As g_k is a power of β , $\nu(g_k) = 0$. Thus, it suffices to express $\nu(g_j)$ in terms of $\nu(g_{j+1})$.

We first express $\nu(g_0)$ in terms of $\nu(g_1)$. Suppose $l_{i_1} = \bar{\alpha}$. Then using Lemma 5.2 and Proposition 2.6, we get

$$\nu(g_0) = \nu(\alpha g_0) = \nu(g_1) + b(w(0, i_1))$$

as g_1 is the result of canceling the first letter of αg_0 with the letter corresponding to l_{i_1} . Similarly, if $l_{i_1} = \alpha$, we get

$$\nu(g_0) = \nu(\bar{\alpha} g_0) = \nu(g_1) - b(w(0, i_1))$$

as can be readily deduced from Lemma 5.2.

We now proceed inductively. We can use the same procedure as above to relate $\nu(g_j)$ with $\nu(g_{j+1})$. As only letters that are α or $\bar{\alpha}$ are deleted, the numbers $b(w(0, i_j))$ are not altered during the inductive construction. Hence we still have

$$\nu(g_j) = \nu(g_{j+1}) \pm b(w(0, i_j))$$

with the sign determined by whether l_{i_j} is α or $\bar{\alpha}$. Using the formula recursively gives the claim. \square

7. A RIGIDITY THEOREM

Consider a word g with $a(g) = b(g) = 0$. Let n_α be the number of letters in g that are α (and hence the number of letters that are $\bar{\alpha}$) and let n_β be the number of letters that are β . Then the number of letters n of g is $2(n_\alpha + n_\beta)$.

Theorem 7.1. *We have $\nu(g) \leq \left(\frac{n}{4}\right)^2$ with equality if and only if $n_\alpha = n_\beta = \frac{n}{4}$ and g is conjugate to the commutator $[\alpha^{n/4}, \beta^{n/4}]$.*

Proof. By Lemma 5.4, there is a complete α -fold \mathcal{F} for g . By Lemma 5.5, we have

$$\nu(g) = \sum_{l=1}^k b(w(i_l, j_l))$$

Observe that as $w(i, j)$ is a sub-word of g , $b(w(i, j)) \leq n_\beta$. The number of terms in the above sum is n_α . Hence it follows that $\nu(g) \leq n_\alpha n_\beta$. As the geometric mean is at most the arithmetic mean and $n = 2(n_\alpha + n_\beta)$, it follows that

$$\nu(g) \leq n_\alpha n_\beta \leq \left(\frac{n}{4}\right)^2$$

In case of equality, each of the above inequalities must be an equality. Hence as the arithmetic mean equals the geometric mean, $n_\alpha = n_\beta = n/4$. Further, for each pair (i, j) , $b(w(i, j)) = n_\beta$. It follows that the letters in $w(i, j)$ include all the letters in g that are β and none of the letters that are $\bar{\beta}$. It is easy to deduce that g is conjugate to $[\alpha^{n/4}, \beta^{n/4}]$. \square

8. THE HEISENBERG INVARIANT AND LOCAL MINIMA

To motivate our next (and most interesting) result, consider the word $[\alpha^{n/4}, \beta^{n/4}]$ (or more generally a word of the form $[\alpha^k, \beta^l]$). It is easy to see that any folding of this word is either an α -folding or a β -folding. Hence to pass from a complete α -folding to a complete β -folding at some intermediate stage, the RNA strand must be completely unfolded.

The word $[\alpha^{n/4}, \beta^{n/4}]$ is characterised by the Heisenberg invariant. However, this situation is too special. We show that if the Heisenberg invariant is close to the maximum value in an appropriate sense, then we have foldings \mathcal{F}_1 and \mathcal{F}_2 so that while passing from \mathcal{F}_1 to \mathcal{F}_2 , at some intermediate stage there are significantly fewer pairs than in \mathcal{F}_1 and \mathcal{F}_2 . To make this precise we introduce some notation.

Fix a word g with $a(g) = b(g) = 0$ and let n , n_α and n_β be as before. For a folding \mathcal{F} of g , we define the deficiency $\rho(\mathcal{F})$ as

$$\rho(\mathcal{F}) = 1 - \frac{2|\mathcal{F}|}{n}$$

This is the fraction of letters that are not in some pair. The potential energy can be assumed to be a monotonically increasing function of the deficiency.

We say that foldings \mathcal{F} and \mathcal{F}' are *adjacent* if their symmetric difference consists of a single pair. A *path* from \mathcal{F} to \mathcal{F}' is a sequence of foldings $\mathcal{F} = \mathcal{F}_0, \mathcal{F}_1, \mathcal{F}_2, \dots, \mathcal{F}_k = \mathcal{F}'$ such that \mathcal{F}_l is adjacent to \mathcal{F}_{l+1} for $0 \leq l < k$. This represents a sequence of steps by which an RNA molecule can pass between one folding and another.

We can now state our result. Assume that k is an integer with k/n small. Let \mathcal{F} be a complete α -pairing for g and \mathcal{F}' a complete β -pairing. We show that if $\nu(g)$ is close to maximal, then for any path from \mathcal{F} to \mathcal{F}' , some path has much larger deficiency than both \mathcal{F} and \mathcal{F}' .

Theorem 8.1. *Suppose $\nu(g) \geq \left(\frac{n}{4}\right)^2 - k^2$. Then,*

- (1) $\rho(\mathcal{F}) \leq \frac{1}{2} + \frac{2k}{n}$ and $\rho(\mathcal{F}') \leq \frac{1}{2} + \frac{2k}{n}$
- (2) *Given any path $\mathcal{F}_0 = \mathcal{F}, \mathcal{F}_1, \mathcal{F}_2, \dots, \mathcal{F}_k = \mathcal{F}'$ from \mathcal{F} to \mathcal{F}' , for some l , $1 \leq l < k$, $\rho^2(\mathcal{F}_l) \geq \frac{1}{2} - 24\left(\frac{k}{n}\right)^2$.*

Remark 8.2. If we assume that k/n is small, then Theorem 8.1 says that deficiency of the foldings \mathcal{F} and \mathcal{F}' are not much more than $1/2$, while that of some intermediate folding is not much less than $1/\sqrt{2}$. Thus, the deficiency, and hence the potential energy, increases significantly in passing from \mathcal{F} to \mathcal{F}' .

Proof of Theorem 8.1. Assume henceforth that the hypothesis of Theorem 8.1 is satisfied. Our first step is as in Theorem 7.1.

Lemma 8.3. *We have $n_\alpha \geq n/4 - k$ and $n_\beta \geq n/4 - k$.*

Proof. Let $n_\alpha = n/4 - p$ with p an integer. Then $n_\beta = n/4 + p$. As $a(g) = b(g) = 0$, applying Theorem 5.5 for the folding \mathcal{F} , we get

$$\nu(g) = \sum_{l=1}^k b(w(i_l, j_l))$$

As \mathcal{F} is a complete α -fold, the number of terms of the above sum is $n/4 - p$. As each word $w(i, j)$ is a sub-word of g , $b(w(i, j)) \leq n_\beta = n/4 + p$. Thus,

$$\nu(g) \leq \left(\frac{n}{4} - p\right) \left(\frac{n}{4} + p\right) = \left(\frac{n}{4}\right)^2 - p^2$$

As $\nu(g) \geq \left(\frac{n}{4}\right)^2 - k^2$, $p^2 \leq k^2$, from which the lemma follows. \square

As \mathcal{F} is a complete α -pairing, $|\mathcal{F}| = n_\alpha$. Hence, as $n_\alpha \geq n/4 - k$, an easy calculation shows that $\rho(\mathcal{F}) \leq \frac{1}{2} + \frac{2k}{n}$ as claimed. Similarly, $\rho(\mathcal{F}') \leq \frac{1}{2} + \frac{2k}{n}$.

We now turn to the second part of the Theorem. We shall first make some observations. Let \mathcal{F}'' be a folding of g with m_α pairs involving α and m_β pairs involving β . We shall use the formula of Lemma 5.3, namely

$$\nu(g) = \nu(g_k) + \sum_{l=1}^k c(i_l, j_l)$$

Consider first the sum $\sum_{l=1}^k c(i_l, j_l)$. This has two kinds of terms corresponding to α -pairs and β -pairs. We consider these separately. First consider the m_α -terms corresponding to $(i, j) \in \mathcal{F}''$ with $l_i = \alpha$. Then $c(i, j) = b(w(i, j))$.

The key observation is that $b(w(i, j)) \leq n_\beta - m_\beta$. This is because $b(w(i, j))$ is the difference between the number of letters in $w(i, j)$ that are β and the number that are $\bar{\beta}$. As there is no nesting, if $(i', j') \in \mathcal{F}''$ with $l_{i'} = \beta$ contained in $w(i, j)$, then $l_{j'} = \bar{\beta}$ is also contained in $w(i, j)$. Hence the net contribution of the letters of the pair to $b(w(i, j))$ is zero.

Thus, the sum of the terms corresponding to pairs (i, j) with $l_i = \alpha$ is at most $m_\alpha(n_\beta - m_\beta)$. We have a similar result for the β -pairs.

Now consider a path as in the hypothesis of the Theorem. It is easy to see that for some l , the number of α -pairs in \mathcal{F}_l is equal to the number of β -pairs. Denote this number by m . We shall find a lower bound for $\rho(\mathcal{F}_l)$. Let the number of elements that are not in any pair be d . If g_k is as above, then g_k has d elements.

Consider the equation

$$(4) \quad \nu(g) = \nu(g_k) + \sum_{l=1}^k c(i_l, j_l)$$

for \mathcal{F}_l . As above, we consider separately the terms corresponding to α -pairs and β -pairs. Let p be as in the proof of Lemma 8.3. We have seen that the total contribution of the α -pairs is at most $m(n_\beta - m) = m(n/4 + p - m)$. By the inequality between arithmetic and geometric means,

$$m(n/4 + p - m) \leq \frac{1}{4} \left(\frac{n}{4} + p\right)^2$$

Similarly, the contribution of the β -terms can be bounded by

$$m(n/4 - p - m) \leq \frac{1}{4} \left(\frac{n}{4} - p\right)^2$$

Hence we get an upper bound on the sum

$$(5) \quad \sum_{l=1}^k c(i_l, j_l) \leq \frac{1}{2} \left(\left(\frac{n}{4}\right)^2 + k^2 \right)$$

using $p^2 \leq k^2$ as in the proof of Lemma 8.3.

By hypothesis, $\nu(g) \geq \left(\frac{n}{4}\right)^2 - k^2$. Hence, from Equation 4 and Equation 5 we get

$$(6) \quad \nu(g_k) \geq \frac{1}{2} \left(\left(\frac{n}{4}\right)^2 - 3k^2 \right)$$

By Theorem 7.1, $\nu(g_k) \leq (d/4)^2$, where d is the number of unpaired letters. By the definition of deficiency, $d = n\rho(\mathcal{F}_l)$. Thus, from Equation 6 we obtain the inequality

$$\left(\frac{n\rho(\mathcal{F}_l)}{4}\right)^2 \geq \frac{1}{2} \left(\left(\frac{n}{4}\right)^2 - 3k^2 \right)$$

or

$$\rho^2(\mathcal{F}_l) \geq \frac{1}{2} - 24 \left(\frac{k}{n}\right)^2$$

as claimed. □

One does not expect in general for the condition $a(g) = b(g) = 0$ to be satisfied. However, one can use Theorem 8.1 provided $a(g)$ and $b(g)$ are small compared to n and $\nu(g) \geq \left(\frac{n}{4}\right)^2 - k^2$ with k/n small. To do this, consider the word $g' = \alpha^{-a(g)}g\beta^{-b(g)}$. By Proposition 2.6, $\nu(g') = \nu(g)$.

The word g' has length $n' = n + a(g) + b(g)$. The hypothesis $\nu(g) \geq \left(\frac{n}{4}\right)^2 - k^2$ can be rephrased as

$$\nu(g') \geq \left(\frac{n'}{4}\right)^2 - k'^2$$

with

$$k'^2 = k^2 + \frac{n'(a(g) + b(g))}{8} - \left(\frac{a(g) + b(g)}{4}\right)^2$$

If k , $a(g)$ and $b(g)$ are all small compared to n , then it follows that k' is small compared to n' (as well as compared to n). Hence we can apply Theorem 8.1 in this case (to the element g' and deduce for g).

We can see that generically $a(g)$ and $b(g)$ are comparable to \sqrt{n} . Namely, a and b for a random string can be viewed as the results of (independent) one-dimensional random walks with n_α and n_β steps. Hence $a(g)$ and $b(g)$ are generically of the order of $\sqrt{n_\alpha}$ and $\sqrt{n_\beta}$, respectively.

Results analogous to those of this section continue to hold if we modify our model so that nearby bases do not pair (as is the case biologically). As further restrictions only increase the number of unpaired bases (in particular of intermediate stages), it suffices to give lower bounds for the number of paired bases (hence upper bounds for the number of unpaired bases) for appropriate α -foldings and β -foldings. An examination of our proof yields such a bound. Namely, if the Heisenberg invariant is close to its maximal value $(n/4)^2$, then (in a sense that can be made precise) for a complete α -folding most terms $b(w(i_l, j_l))$ in Lemma 5.5 must be close to $n/4$. This means that most bonds in the complete α -folding are between bases that are not close to each other. Hence we can obtain a lower bound on the number of bonds in an α -folding without nearby bases paired. The case of β -foldings is similar.

9. LOWER BOUNDS ON DEFICIENCY

We now turn to another important application of the Heisenberg invariant. Let g be a word in the free group F with $a(g) = b(g) = 0$. As the Heisenberg invariant is a measure of non-triviality, one expects that if $\nu(g)$ is large then the deficiency of any folding \mathcal{F} of g is large. We now prove such a result.

Theorem 9.1. *For any folding \mathcal{F} of g , if $\rho = \rho(\mathcal{F})$*

$$\rho - \frac{3\rho^2}{4} \geq \frac{4\nu(g)}{n^2}$$

Proof. As before, we use the formula of Lemma 5.3, namely

$$\nu(g) = \nu(g_k) + \sum_{l=1}^k c(i_l, j_l)$$

Let $d = n\rho(g)$ be the number of letters that are unpaired in \mathcal{F} (hence the number of letters of g_k). We claim that $c(i_l, j_l) \leq d/2$. This follows if the number of unpaired letters of the sub-word $w(i_l, j_l)$ is at most $d/2$ as pairs have canceling contributions to $a(w(i_l, j_l))$ and $b(w(i_l, j_l))$ (as in the Proof of Theorem 8.1). Otherwise $w(j_l, i_l)$ has at most $d/2$ unpaired letters, from which we can deduce the result as $a(w(j_l, i_l)) = -a(w(i_l, j_l))$ and $b(w(j_l, i_l)) = -b(w(i_l, j_l))$ since $a(g) = b(g) = 0$ and l_{i_l} and l_{j_l} are a canceling pair.

The number of pairs in \mathcal{F} is $(n - d)/2$. Further, by Theorem 7.1, $\nu(g_k) \leq (\frac{d}{4})^2$. Hence, as $d = n\rho(g)$, we get

$$\nu(g) \leq \left(\frac{d}{4}\right)^2 + \frac{d(n-d)}{4} = \frac{n^2}{4} \left(\rho(\mathcal{F}) - \frac{3\rho^2}{4}\right)$$

from which the claim follows. \square

10. THE MILNOR INVARIANTS AND OTHER EXTENSIONS

We now turn to the higher Milnor invariants. We shall be very sketchy in this section as our goal is to indicate further extensions of our methods.

We begin by recalling the conceptual scheme for defining the Milnor invariants. Suppose g is an element of the free group F . The Milnor invariants (in our situation) are measures of how different g is from the identity. Further, these are invariant under conjugacy and are additive. Thus, they are very well suited for studying RNA folding.

The Milnor invariants are defined in terms of the lower central series. The first Milnor invariants correspond to the image of g in the abelianisation F/F_2 . If this is trivial, then g is in F_2 , and we consider its image in F_2/F_3 . This is the Heisenberg invariant.

We proceed inductively, with the Milnor invariants of order n defined if those of lower order vanish. Namely, if the invariants of order less than n vanish, then $g \in F_n$. We consider the image of g in F_n/F_{n+1} , which is a finitely generated free abelian group. By choosing a basis for this group, we get finitely many integers determining the image. Thus, we have an infinite series of invariants. It follows from the Magnus expansion homomorphism (which we recall below) that $\bigcap_{i=1}^{\infty} F_i = \{1\}$, hence all the Milnor invariants of g vanish if and only if g is trivial.

In our situation, we would like to define the Milnor invariants of order n up to an error corresponding to the lower order Milnor invariants, as we did in the case

of the Heisenberg invariant. We can do this as we have a given basis for the free group F . Namely, we choose and fix sections $s_k : F/F_k \rightarrow F$, i.e. functions between the underlying sets of F/F_k and F (not preserving the algebraic structure) so that for the quotient homomorphism $q_k : F \rightarrow F/F_k$, we have $q_k \circ s_k : F/F_k \rightarrow F/F_k$ is the identity. Then for an arbitrary $g \in F$, $g \cdot (s_n \circ q_n(g))^{-1}$ is in F_n , so we can define Milnor invariants by considering the image of $g(s_n \circ q_n(g))^{-1}$ in F_n/F_{n+1} . This is what we did in the case of the Heisenberg invariant here, using the section $(a, b) \mapsto \alpha^a \beta^b$. As we are not studying the higher invariants in detail here, we do not construct explicit sections.

We remark that in the original topological context, there is no canonical basis and hence one cannot make a well-defined choice of section. However, the Milnor invariants are defined modulo those of lower order. Such arithmetical considerations are not likely to be fruitful in the context of RNA.

An explicit description of the Milnor invariants, allowing for efficient computation, can be given in terms of the so called *Magnus expansion homomorphism*. Consider formal power series with integer coefficients in two non-commuting variables X and Y (i.e., where XY is not YX for example). These form a ring $\mathbb{Z}[[X, Y]]$. The Magnus expansion homomorphism M is an injective homomorphism from the free group F to the multiplicative group of invertible elements of $\mathbb{Z}[[X, Y]]$. As is well known, the invertible elements of $\mathbb{Z}[[X, Y]]$ are the formal power series with constant term 1. The Magnus homomorphism is the unique homomorphism such that the image of the generators α and β satisfy

$$M(\alpha) = 1 + X$$

$$M(\beta) = 1 + Y$$

By properties of formal power series, we have

$$M(\bar{\alpha}) = 1 - X + X^2 - X^3 + \dots$$

$$M(\bar{\beta}) = 1 - Y + Y^2 - Y^3 + \dots$$

In general, for a word g , we multiply the images of the letters of g to obtain $M(g)$.

For $g \in F$, the constant term of $M(g)$ is 1. The coefficients of X and Y are $a(g)$ and $b(g)$, and thus determine and are determined by the abelianisation. In particular, the coefficients vanish if and only if $a(g) = b(g) = 0$.

Suppose $a(g) = b(g) = 0$. Then it is known that the term of degree two is of the form $c(XY - YX)$. The Heisenberg invariant is $\nu(g) = c$. More generally, $g \in F_n$ if and only if the terms of degree less than n (except the constant term) vanish. As a consequence, we have a well-defined homomorphism of abelian groups from F_n/F_{n+1} to homogeneous polynomials of degree n in the non-commuting variables X and Y . This is injective but not surjective. The image consists of so called *Lie elements*, for which an explicit basis can be given.

Thus, Milnor invariants can be defined as linear combinations of coefficients of the polynomial $M(g)$. This expression is not unique as the Lie elements do not generate all the homogeneous polynomials of degree n . For instance, the Heisenberg invariant can be defined as the coefficient of XY or the negative of the coefficient of YX . If g is in F_n , then all these expressions give the same value. For a general element g , the choice of a linear combination is analogous to the choice of a section s_n in the earlier description. As in the case of the sections s_n , we do not give details of the relevant bases and coefficients.

We finally turn to the original topological context which motivated this work. A *link* L in a smooth 3-dimensional manifold is a disjoint union of smoothly embedded circles. An *unlink* is a link L so that the components of L bound disjoint, smoothly embedded discs. There are three well studied relations on links - those of isotopy, homotopy and concordance. We consider links up to these relations.

Firstly, let L' be an unlink with two components. The fundamental group of the complement is a free group on two generators, and hence can be identified with F . Given a word g in F , consider a three-component link L , with the third component a curve γ representing the element g (up to conjugation) in the complement of L' . This is not well-defined, but we may study properties of this link that depend only on g .

The first observation is that the link is defined up to *link homotopy*, i.e., changing the link through a family so that each component is allowed to cross itself but not others. For such three-component links, the link is determined by exactly three invariants. These correspond to $a(g)$, $b(g)$ and $\nu(g)$. We get further invariants by observing that we can keep the first two components fixed and only allow γ to cross itself. We expect that it will be fruitful to study the link L up to concordance (which implies link homotopy). Link homotopy, isotopy and concordance have been extensively studied. We refer to [1]–[11] for some of the fundamental results concerning these.

11. CONCLUDING REMARKS

In this article, we have constructed an easily computed number, the Heisenberg invariant, associated to a strand of RNA, and showed that it is related to a dynamically important property of Watson-Crick pairing. Values of the invariant large enough to yield dynamically interesting results are not generic. However, in biological systems it is important to understand molecules with special properties, as evolution ensures that living systems are not generic but have compositions giving desirable properties.

We end with a conjecture regarding the Milnor invariants of order one more than the Heisenberg invariant. We have seen that the Heisenberg invariant being large implies that we have minima that are widely separated. We expect that the next invariants being large implies that there are minimax paths between minima that are widely separated. This is significant in the context of catalysis.

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