second binding site, neomycin stabilizes a ribosomal intermediate hybrid state, between locked and unlocked state, impairing translocation and ribosome recycling (Wang et al. 2012).

The aberrant proteins produced are thought to interrupt the cell membrane by creating membrane channels, further allowing AGAs to penetrate the cell wall. This rapid uptake of AGAs leads eventually to lethality (Davis et al. 1986).

1.2 Mechanisms of Resistance

Bacterial resistance to AGAs is the result of a combined effect of three main causes (highlighted in the following subsections): the action of aminoglycoside-modifying enzymes (AMEs) (Section 1.2.1), the mutation and modification of AGA's target sequence in the ribosome (Section 1.2.2), and changes in uptake and efflux (Section 1.2.3). However, other causes may have a major influence in pathogenic organisms, such as in *Pseudomonas aeruginosa*: on the one hand, membrane proteases counteract the effect of aminoglycosides by eliminating the misfolded/mistranslated proteins produced in aminoglycoside-affected ribosomes (Kindrachuk et al. 2011; Krahn et al. 2012); on the other hand, biofilm production can impair the access of AGAs to the bacterial cells (Poole 2011). Moreover, *P. aeruginosa* has been found to produce periplasmic cyclic β-glucans, capable of directly binding kanamycin (Sadovskaya et al. 2010).

1.2.1 Aminoglycoside-Modifying Enzymes

The most clinically significant mechanism of aminoglycoside resistance is a consequence of enzymatic modification of specific chemical groups in the antibiotics. Interestingly, it is hypothesized that the enzymes originate from the organisms that produce the aminoglycoside itself (Perry et al. 2014). Over a hundred different enzymes have been identified, and the genes coding for AMEs are usually found on plasmids and transposons. Extensive reviews on the diverse AMEs, organisms, mechanisms of resistance to AGAs, strategies of inhibition, and 3D crystal structures have been done (Ramirez and Tolmasky 2010; Labby and Garneau-Tsodikova 2013; Garneau-Tsodikova and Labby 2016). There are three types of AMEs: (i) ATP (and/or GDP)-dependent aminoglycoside phosphotransferases (APHs), (ii) acetyl-CoA-dependent aminoglycoside acetyltransferases (AACs), and (iii) ATP-dependent aminoglycoside nucleotidyltransferases (ANTs). Figure 1.3 illustrates the action of the three types of enzymes, including the known crystal structures of representative members in complex with aminoglycosides and catalytic cofactors.

As AGAs target specific regions and sequences of RNA with particular orientations, the described enzymatic modifications in the basic structure of