

The gel of alginate is formed by dimerization of chain that is further cross-linked to other chains [25].

10.3.2 Ionic Cross-Linking

Alginate is a copolymer that has distinct affinities against the distinct divalent ions. The stability, permeability, and strength of alginate gel depend on the cation that will be used in the formation of gel [26]. The exchange monovalent cations from the surrounding environment and the divalent cations affect the stability of hydrogel by weakening the mechanical properties [4]. The concentration, source, degree and type of cross-linking, and molecular weight limit the delivery system of drugs, protein agents, and cells [27]. Besides these factors, the physiological conditions, whether they are *in vitro* or *in vivo*, affect the stability. One of the divalent cation Ca^{2+} -based hydrogel alginates is a very less stable cross-linking hydrogel. If 0.9 weight % sodium chloride is added in the solution, there is an exchange of calcium by non-gelling monovalent sodium ions as well as by chelators such as citrate or phosphate present in the solution [28]. The monovalent sodium ions act as de-cross-linking agents. The cell culture medium *in vitro* condition has sufficiently high calcium concentration that counterbalances such kind of effects, but this hydrogel nevertheless remains stable for weeks [29].

10.3.3 External Gelation

Calcium chloride (CaCl_2) is the most common and frequently used soluble salt, which acts as ionic cross-linker agents in external gelation. External gelation is a very transparent process that creates a nontoxic environment for cell entrapment [4]. However, although gelation occurs almost instantaneously, this process frequently results in unbalanced cross-linking density and a polymer concentration gradient within the formed hydrogel [30]. Gel beads are formed by exuding a sodium alginate solution into an aqueous solution of calcium ions. Generally, these microbeads are produced by coaxial airflow that controls the size of the droplets by blowing them from a needle tip into a CaCl_2 bath. In tissue regeneration, cell delivery system alginate beads have been widely used [31–33].

The size of microbeads is dependent on the flow of air and the diameter of the solution. The electrostatic bead generation technique is used to attain smaller size beads up to 150 μm [34]. For the production of spherical hydrogels, a novel method was used that utilizes super hydrophobic substrates. Without any precipitation, the spherical hydrogels entangle